These records are from CDER's historical file of information previously disclosed under the Freedom of Information Act (FOIA) for this drug approval and are being posted as is. They have not been previously posted on Drugs@FDA because of the quality (e.g., readability) of some of the records. The documents were redacted before amendments to FOIA required that the volume of redacted information be identified and/or the FOIA exemption be cited. These are the best available copies.

N 50580 -1

NDA 50-580

AP/LTR

NDA 50-580

Norman W. Lavy, M.D.
E.R. Squibb & Sons, Inc.
P.O. Box 191
New Brunswick, NJ 08903

Dear Dr. Lavy:

Reference is made to your New Drug Application dated June 1, 1983 submitted pursuant to section 505(b) of the Federal Food, Drug and Cosmetic Act for Azactam (aztreonam) for Injection.

We also acknowledge receipt of your additional communications dated October 20, November 30, 1983; January 5, 16, 17, February 1, 15, April 19, 23, July 12, 16, August 16, 22, 30, September 10, 18, October 3, 10, November 26, and December 28, 1984; January 23, February 27, April 1, 11, May 9, 13, June 7, July 16, September 10, November 13, December 16, 1985 and January 10, 13, 28, February 10, 13, 24, October 28, November 6, December 10, 17, 21, 24, 29, 30, 1986.

We have completed review of this application and have concluded that adequate information has been presented to demonstrate that the drug is safe and effective as recommended in the final printed labeling numbered J4-140 submitted on December 30, 1986. Accordingly, the application is approved, effective on the date of this letter.

Please submit, in duplicate, the advertising copy which you intend to use in your proposed introductory promotional and/or advertising campaign. Please submit one copy to the Division of Anti-Infective Drug Products, and the second copy to the Division of Drug Advertising and Labeling, HFN-240, Room 10B-04, 5600 Fishers Lane, Rockville, Maryland 20857. Please submit all proposed materials in draft or mock-up form, not final print. Also, please do not use form FD-2253 for this submission; this form is for routine use, not proposed material.

Please submit one market package of the drug when available.

We remind you that you must comply with the requirements set forth under CFR 314.80 and 314.81 for an approved NDA.

Sincerely yours,

12/31/86

Elaine C. Esber, M.D.
Director
Office of Biologics Research and Review
Center for Drugs and Biologics

cc:
Orig NDA 50-580
NWK-DO
HFN-82
HFN-220
HFN-535
HFN-710
HFN-800/JMinor
HFN-815 12/30/86
HFN-815/CSO/KCreedon/12/30/86/11m/1887m
HFN-815/MO/GStanley
HFN-815/PHARM/.../JDavitt
HFN-815/MICRO/RNorton/PDionne

50-580

AE LTR

NDA 50-580

Norman W. Lavy, M.D.
E.R. Squibb & Sons, Inc.
P.O. Box 191
New Brunswick, NJ 08903

Dear Dr. Lavy:

Reference is made to your New Drug Application (NDA) for Aztreonam (monobactam) for Injection.

We have completed our review of this application, and it is approvable for the following indications:

1. Urinary tract infections (complicated and uncomplicated), including pyelonephritis and cystitis (initial and recurrent) caused by Escherichia coli, Klebsiella pneumoniae, Proteus mirabilis, Pseudomonas aeruginosa, Enterobacter cloacae, Klebsiella oxytoca*, Citrobacter species* and Serratia marcescens*.

2. Lower respiratory tract infections, including pneumonia and bronchitis caused by E. coli, K. pneumoniae, P. aeruginosa, Haemophilus influenzae, P. mirabilis, Enterobacter species and S. marcescens*.

3. Gynecologic infections, including endometritis and pelvic cellulitis caused by E. coli, K. pneumoniae*, Enterobacter species* including E. cloacae* and P. mirabilis*.

4. Intra-abdominal infections, including peritonitis caused by E. coli, Klebsiella species including K. pneumoniae, Enterobacter species including E. cloacae*, P. aeruginosa, Citrobacter species* including C. freundii* and Serratia species* including S. marcescens*.

5. Skin and skin-structure infections, including those about tendons, infected postoperative wounds, ulcers and burns caused by E. coli, P. mirabilis, S. marcescens, Enterobacter species, P. aeruginosa, K. pneumoniae, and Citrobacter species*.

6. Septicemia caused by E. coli, K. pneumoniae, P. aeruginosa, P. mirabilis*, S. marcescens* and Enterobacter species.

*Efficacy for this organism in this organ system was studied in fewer than 10 infections.

The use of azithromycin [illegible] because the total number of female patients was [illegible] approval.

The single-dose intracellular [illegible] urinary tract infection (cystitis) [illegible] multicenter, randomized [illegible] therapy is less effective than the conventional [illegible].

The revised draft package insert dated [illegible], 1991 (copy attached) should incorporate only the above claims and the following changes:

1. Under CLINICAL PHARMACOLOGY:

   a. In the thirteenth [illegible] sentences, "Twenty patients with [illegible] AZAC PAM." Lack of reactivity to skin test or lack of antibodies are not [illegible] that a penicillin reaction was not absent. The labeling should not imply [illegible] guarantee for lack of an [illegible] reaction can be proven in large populations.

   b. In the [illegible] sentence of [illegible] "[illegible] appropriate intestinal [illegible].

2. Under [illegible]:

   a. Indications other [illegible] this letter, should be deleted.

   b. Under Urinary Tract Infections, [illegible] should be deleted.

   c. Under Gynecologic Infections, "pelvic [illegible] should be deleted.

   d. The paragraph "A&A[illegible] has proven [illegible]" should be deleted.

   e. In the Concurrent [illegible] begins "Patients may [illegible]".

3. Under WARNINGS the original proposed wording, "Careful study should be made..." should be retained.

4. Under General Precautions:

   a. The first paragraph should be deleted.

   b. The first sentence of the fourth paragraph should read: "The use of antibiotics may promote the overgrowth of nonsusceptible organisms, including gram-positive organisms (Staphylococcus aureus and Streptococcus faecalis) and fungi."

5. Under ADVERSE REACTIONS:

   a. Under Adverse laboratory changes, the statement about hepatic change should read: hepatic - elevations of AST (SGOT), ALT (SGPT), and alkaline phosphatase; signs or symptoms of hepatobiliary dysfunction occurred in less than 1% of recipients (see above).

6. Under DOSAGE AND ADMINISTRATION:

   a. The third paragraph starting "A similar [illegible]..." should be deleted.

   b. The fifth paragraph starting "For [illegible] of other antibiotics ..." should be deleted.

Before the application can be approved, revised labeling must be submitted, and a safety update report must be submitted in accordance with 314.50(d)(5)(vi)(b).

Within 10 days after the date of this letter, you are required to amend the application, or notify us of your intent to file an amendment, or follow one of the other options under 21 CFR 314.110. In the absence of such action, the Food and Drug Administration may take action to withdraw the application.

NDA 50-560
Page 4

The drug may not be legally marketed until you have been notified in writing that the application is approved.

Sincerely yours,

for Robert A. Rubin 10/16/86

Elaine S. Tabor, M.D.
Director
Office of Biologics Research and Review
Center for Drugs and Biologics

Enclosure
cc: NWK-DO
ORIG. NDA 50-560 GRS on Label
HFN-82 10/16/86
HFN-220
HFN-535
HFN-710
HFN-800/JMinor
HFN-815
HFN-815/DSO/FCrecelius/12/11/85/SAJ/12/15/85/10/14/86
HFN-815/MO/FMin/5/6/86/GRStanley/5/7/86
HFN-815/PHARM/SNAlam/4/29/86/JDavitt/4/30/86
HFN-815/MICRO/RNorton/4/29/86/5/1/86/PDionne/4/29/86
R/D init. by: ETabor/6/10/86/6/20/86
F/D: 4/28/86/5/2/86/5/30/86/6/16/86/6/26/86/10/14/86
F/T: 6/26/86/7/1/86/10/14/86

APPROVABLE 0204u

OAA 10-15-86

FPL

SQUIBB

J4-140

APPROVED
DEC 3 1 1981

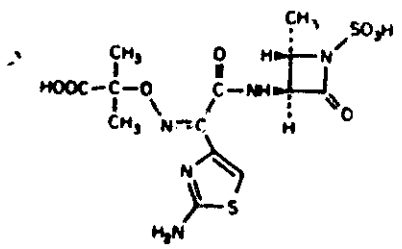
CAUTION: Federal law prohibits dispensing without prescription.

AZACTAM® FOR INJECTION
Aztreonam For Injection

DESCRIPTION
AZACTAM (aztreonam, Squibb) is the first member of a new class of antibiotics developed by the Squibb Institute for Medical Research and classified as monobactams. These agents were originally isolated from *Chromobacterium violaceum*. AZACTAM is a totally synthetic bactericidal antibiotic with activity against a wide spectrum of gram-negative aerobic pathogens.

The monobactams, having a unique monocyclic beta-lactam nucleus, are structurally different from other beta-lactam antibiotics (e.g., penicillins, cephalosporins, cephamycins). The sulfonic acid substituent in the 1-position of the ring activates the beta-lactam moiety; an aminothiazolyl oxime side chain in the 3-position and a methyl group in the 4-position confer the specific antibacterial spectrum and beta-lactamase stability.

Aztreonam is designated chemically as (Z)-2-[[[(2-amino-4-thiazolyl)[[(2S,-3S)-2-methyl-4-oxo-1-sulfo-3-azetidinyl]carbamoyl]methylene]amino]oxy]-2-methylpropionic acid. Structural formula:



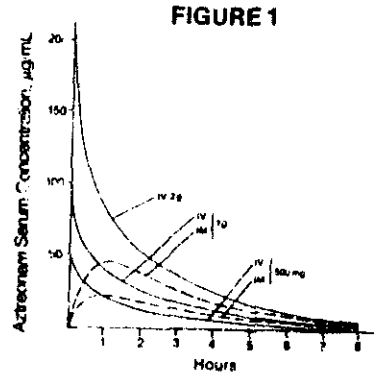
$C_{13}H_{17}N_5O_8S_2$ MW 435.42 CAS-78110-38-0

AZACTAM For Injection (Aztreonam For Injection) is a sterile nonpyrogenic white powder, containing approximately 780 mg L-arginine per gram of aztreonam, for intramuscular or intravenous use following constitution. The powder is sodium-free. Aqueous solutions of aztreonam have a pH in the range of 4.5 to 7.5.

CLINICAL PHARMACOLOGY
Single 30-minute intravenous infusions of 500 mg, 1 g and 2 g doses of AZACTAM in healthy subjects produced peak serum levels of 54, 90 and 204 µg/mL, respectively, immediately after administration; at eight hours, serum levels were 1, 3 and 6 µg/mL, respectively (Figure 1). Single 3-minute intravenous injections of the same doses resulted in serum levels of 58, 125 and 242 µg/mL at five minutes following completion of injection.

Serum concentrations of aztreonam in healthy subjects following completion of single intramuscular injections of 500 mg and 1 g doses are depicted in Figure 1; maximum serum concentrations occur at about one hour. After identical single intravenous or intramuscular doses of AZACTAM, the serum concentrations of aztreonam are comparable at one hour (1.5 hours from start of intravenous infusion) with similar slopes of serum concentrations thereafter.

FIGURE 1



The serum levels of aztreonam following single 500 mg, or 1 g (intramuscular or intravenous) or 2 g (intravenous) doses of AZACTAM (aztreonam) exceed the MIC_{90} for *Neisseria* sp., *H. influenzae* and most genera of the *Enterobacteriaceae* for eight hours (for *Enterobacter* sp., the eight hour serum levels exceed the MIC for 80 percent of strains). For *Ps. aeruginosa*, a single 2 g intravenous dose produces serum levels that exceed the MIC_{90} for approximately four to six hours. All of the above doses of AZACTAM result in average urine levels of aztreonam that exceed the MIC_{90} for the same pathogens for up to 12 hours.

The serum half-life of aztreonam averaged 1.7 hours (1.5 to 2.0) in subjects with normal renal function, independent of the dose and route of administration. In healthy subjects, based on a 70 kg person, the serum clearance was 91 mL/min and renal clearance was 56 mL/min; the apparent mean volume of distribution at steady-state averaged 12.6 liters, approximately equivalent to extracellular fluid volume.

In a study of healthy elderly male subjects (65 to 75 years of age), the average elimination half-life of aztreonam was slightly longer than in young healthy males.

In patients with impaired renal function, the serum half-life of aztreonam is prolonged (see DOSAGE AND ADMINISTRATION, Renal Impairment). The serum half-life of aztreonam is only slightly prolonged in patients with hepatic impairment since the liver is a minor pathway of excretion.

Average urine concentrations of aztreonam were approximately 1100, 3500 and 6600 µg/mL within the first two hours following single 500 mg, 1 g and 2 g intravenous doses of AZACTAM (30-minute infusions), respectively. The range of average concentrations for aztreonam in the 8 to 12 hour urine specimens in these studies was 25 to 120 µg/mL. After intramuscular injection of single 500 mg and 1 g doses of AZACTAM, urinary levels were approximately 500 and 1200 µg/mL, respectively, within the first two hours, declining to 180 and 470 µg/mL in the six to eight hour specimens. In healthy subjects, aztreonam is excreted in the urine about equally by active tubular secretion and glomerular filtration. Approximately 60 to 70 percent of an intravenous or intramuscular dose was recovered in the urine by eight hours. Urinary excretion of a single parenteral dose was essentially complete by 12 hours after injection. About 12 percent of a single intravenous radiolabeled dose was recovered in the feces. Unchanged aztreonam and the inactive beta-lactam ring hydrolysis product of aztreonam were present in feces and urine.

Intravenous or intramuscular administration of a single 500 mg or 1 g dose of AZACTAM (aztreonam) every eight hours for seven days to healthy subjects produced no apparent accumulation of aztreonam or modification of its disposition characteristics, serum protein binding averaged 56 percent and was independent of dose. An average of about 6 percent of a 1 g intramuscular dose was excreted as a microbiologically inactive open beta-lactam ring hydrolysis product (serum half-life approximately 28 hours) of aztreonam in the zero to eight hour urine collection on the last day of multiple dosing.

Renal function was monitored in healthy subjects given aztreonam; standard tests (serum creatinine, creatinine clearance, BUN, urinalysis and total urinary protein excretion) as well as special tests (excretion of N-acetyl-β-glucosaminidase, alanine aminopeptidase and β_2-microglobulin) were used. No abnormal results were obtained.

SQUIBB

J4-140

APPROVED
DEC 31 1987

CAUTION: Federal law prohibits dispensing without prescription.

AZACTAM® FOR INJECTION
Aztreonam For Injection

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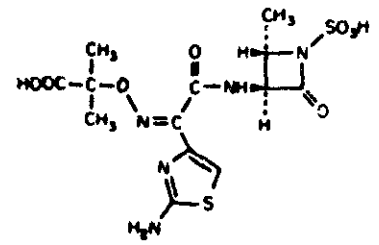

$C_{13}H_{17}N_5O_8S_2$ MW 435.42 CAS-78110-38-0

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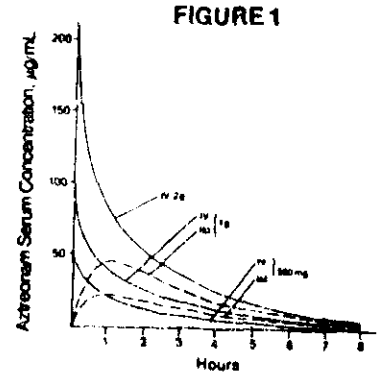
FIGURE 1



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Intravenous or intramuscular administration of a single 500 mg or 1 g dose of AZACTAM (aztreonam) every eight hours for seven days to healthy subjects produced no apparent accumulation of aztreonam or modification of its disposition characteristics; serum protein binding averaged 56 percent and was independent of dose. An average of about 8 percent of a 1 g intramuscular dose was excreted as a microbiologically inactive open beta-lactam ring hydrolysis product (serum half-life approximately 26 hours) of aztreonam in the zero to eight hour urine collection on the last day of multiple dosing.

Renal function was monitored in healthy subjects given aztreonam; standard tests (serum creatinine, creatinine clearance, BUN, urinalysis and total urinary protein excretion) as well as special tests (excretion of N-acetyl-β-glucosaminidase, alanine aminopeptidase and β_2-microglobulin) were used. No abnormal results were obtained.

Aztreonam achieves measurable concentrations in the following body fluids and tissues:

EXTRAVASCULAR CONCENTRATIONS OF AZTREONAM AFTER A SINGLE PARENTERAL DOSE[1]

Fluid or Tissue	Dose (g)	Route	Hours Postinjection	Number of Patients	Mean Concentration (μg/mL or μg/g)
Fluids					
bile	1	IV	2	10	39
blister fluid	1	IV	1	6	20
bronchial secretion	2	IV	4	7	5
cerebrospinal fluid (inflamed meninges)	2	IV	0.9-4.3	16	3
pericardial fluid	2	IV	1	6	33
pleural fluid	2	IV	1.1-3.0	3	51
synovial fluid	2	IV	0.8-1.9	11	83
Tissues					
atrial appendage	2	IV	0.9-1.6	12	22
endometrium	2	IV	0.7-1.9	4	9
fallopian tube	2	IV	0.7-1.9	8	12
fat	2	IV	1.3-2.0	10	5
femur	2	IV	1.0-2.1	15	16
gallbladder	2	IV	0.8-1.3	4	23
kidney	2	IV	2.4-5.6	5	87
large intestine	2	IV	0.8-1.9	9	12
liver	2	IV	0.9-2.0	6	47
lung	2	IV	1.2-2.1	6	22
myometrium	2	IV	0.7-1.9	9	11
ovary	2	IV	0.7-1.9	7	13
prostate	1	IM	0.8-3.0	8	8
skeletal muscle	2	IV	0.3-0.7	6	16
skin	2	IV	0.0-1.0	8	25
sternum	2	IV	1	6	6

[1]Tissue penetration is regarded as essential to therapeutic efficacy, but specific tissue levels have not been correlated with specific therapeutic effects.

The concentration of aztreonam in saliva at 30 minutes after a single 1 g intravenous dose (9 patients) was 0.2 μg/mL; in breast milk at two hours after a single 1 g intravenous dose (6 patients), 0.2 μg/mL, and at six hours after a single 1 g intramuscular dose (6 patients), 0.3 μg/mL; in amniotic fluid at six to eight hours after a single 1 g intravenous dose (5 patients), 2 μg/mL. The concentration of aztreonam in peritoneal fluid obtained one to six hours after multiple 2 g intravenous doses ranged between 12 and 90 μg/mL in 7 of 8 patients studied.

Aztreonam given intravenously rapidly reaches therapeutic concentrations in peritoneal dialysis fluid; conversely, aztreonam given intraperitoneally in dialysis fluid rapidly produces therapeutic serum levels.

Concomitant administration of probenecid or furosemide and AZACTAM (aztreonam) causes clinically insignificant increases in the serum levels of aztreonam. Single-dose intravenous pharmacokinetic studies have not shown any significant interaction between aztreonam and concomitantly administered gentamicin, nafcillin sodium, cephradine, clindamycin or metronidazole. No reports of disulfiram-like reactions with alcohol ingestion have been noted; this is not unexpected since aztreonam does not contain a methyl-tetrazole side chain.

The implications of the following information for predicting the occurrence of hypersensitivity reactions to AZACTAM have not been established. The number of patients included in immunologic studies is too small to draw firm conclusions with regard to clinical practice:

> A study in rabbits suggests that antibodies produced in response to benzylpenicillin and to cephalothin show little cross-reactivity with aztreonam, and antibodies produced in response to aztreonam show little cross-reactivity with benzylpenicillin and cephalothin.
>
> In a group of 22 subjects with positive skin tests to penicillin reagents, three also had positive skin tests to aztreonam. One was negative on retesting, one was confirmed as positive, and the third subject refused further evaluation. The 20 subjects with negative aztreonam skin tests were given one injection of AZACTAM 1 g IM. There were no immediate hypersensitivity reactions, but one subject later developed a localized rash that was compatible with a fixed drug eruption.
>
> In 36 subjects receiving multiple doses of AZACTAM over a seven-day period, no IgE antibody response was detectable and only one subject demonstrated an IgG response.

Microbiology

Aztreonam exhibits potent and specific activity *in vitro* against a wide spectrum of gram-negative aerobic pathogens including *Pseudomonas aeruginosa*. The bactericidal action of aztreonam results from the inhibition of bacterial cell wall synthesis due to a high affinity of aztreonam for penicillin binding protein 3 (PBP3). Aztreonam, unlike the majority of beta-lactam antibiotics, does not induce beta-lactamase activity and its molecular structure confers a high degree of resistance to hydrolysis by beta-lactamases (i.e., penicillinases and cephalosporinases) produced by most gram-negative and gram-positive pathogens. It is therefore usually active against gram-negative aerobic organisms that are resistant to antibiotics hydrolyzed by beta-lactamases. Aztreonam maintains its antimicrobial activity over a pH range of 6 to 8 *in vitro*, as well as in the presence of human serum and under anaerobic conditions. Aztreonam is active *in vitro* and is effective in laboratory animal models and clinical infections against most strains of the following organisms, including many that are multiply-resistant to other antibiotics (i.e., certain cephalosporins, penicillins, and aminoglycosides):

Escherichia coli
Enterobacter species
Klebsiella species, including *K. pneumoniae* and *K. oxytoca*
Proteus mirabilis
Pseudomonas aeruginosa
Serratia marcescens
Haemophilus influenzae (including ampicillin-resistant and other penicillinase-producing strains)
Citrobacter species

While *in vitro* studies have demonstrated the susceptibility to aztreonam of most strains of the following organisms, clinical efficacy for infections other than those included in the INDICATIONS AND USAGE section has not been documented:

Neisseria gonorrhoeae (including penicillinase-producing strains)
Proteus vulgaris
Morganella morganii (formerly *Proteus morganii*)
Providencia species, including *P. stuartii* and *P. rettgeri* (formerly *Proteus rettgeri*)
Pseudomonas species
Shigella species
Pasteurella multocida
Yersinia enterocolitica
Aeromonas hydrophila
Neisseria meningitidis

Aztreonam and aminoglycosides have been shown to be synergistic *in vitro* against most strains of *Ps. aeruginosa*, many strains of *Enterobacteriaceae*, and other gram-negative aerobic bacilli.

Alterations of the anaerobic intestinal flora by broad spectrum antibiotics may decrease colonization resistance, thus permitting overgrowth of potential pathogens, e.g., *Candida* and Clostridia species. Aztreonam has little effect on the anaerobic intestinal microflora in *in vitro* studies. *Clostridium difficile* and its cytotoxin were not found in animal models following administration of aztreonam (see ADVERSE REACTIONS, Gastrointestinal).

Susceptibility Testing

Diffusion Technique: Quantitative procedures that require measurement of zone diameters give precise estimates of microbial susceptibility to antibiotics. One such method, recommended for use with the aztreonam 30 μg disk, is the National Committee of Clinical Laboratory Standards (NCCLS) approved procedure. Only a 30 μg aztreonam disk should be used; there are no suitable surrogate disks.

Results of laboratory tests using 30 μg aztreonam disks should be interpreted using the following criteria:

Zone Diameter (mm)	Interpretation
≥ 22	(S) Susceptible
16 - 21	(I) Intermediate (Moderate Susceptibility)
≤ 15	(R) Resistant

Dilution Technique: Broth or agar dilution methods may be used to determine the minimal inhibitory concentration (MIC) of aztreonam.

MIC test results should be interpreted according to the concentrations of aztreonam that can be attained in serum, tissues and body fluids.

MIC (μg/mL)	Interpretation
≤ 8	(S) Susceptible
16	(I) Intermediate (Moderate Susceptibility)
≥ 32	(R) Resistant

For any susceptibility test, a report of "susceptible" indicates that the pathogen is likely to respond to AZACTAM therapy; a report of "resistant" indicates that the pathogen is not likely to respond. A report of "intermediate" (moderate susceptibility) indicates that the pathogen is expected to be susceptible to AZACTAM (aztreonam) if high dosages are used, or if the infection is confined to tissues and fluids (e.g., urine, bile) in which high aztreonam levels are attained.

The quality control cultures should have the following assigned daily ranges for aztreonam:

		Disks	Mode MIC (μg/mL)
E. coli	(ATCC 25922)	28-36 mm	0.06-0.25
Ps. aeruginosa	(ATCC 27853)	23-29 mm	2.0-8.0

INDICATIONS AND USAGE

Before initiating treatment with AZACTAM, appropriate specimens should be obtained for isolation of the causative organism(s) and for determination of susceptibility to aztreonam. Treatment with AZACTAM may be started empirically before results of the susceptibility testing are available; subsequently, appropriate antibiotic therapy should be continued.

AZACTAM For Injection (Aztreonam For Injection) is indicated for the treatment of the following infections caused by susceptible gram-negative microorganisms:

Urinary Tract Infections (complicated and uncomplicated), including pyelonephritis and cystitis (initial and recurrent) caused by *Escherichia coli, Klebsiella pneumoniae, Proteus mirabilis, Pseudomonas aeruginosa, Enterobacter cloacae, Klebsiella oxytoca**, *Citrobacter* species* and *Serratia marcescens**.

Lower Respiratory Tract Infections, including pneumonia and bronchitis caused by *Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Haemophilus influenzae, Proteus mirabilis, Enterobacter* species and *Serratia marcescens**.

Septicemia caused by *Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Proteus mirabilis**, *Serratia marcescens** and *Enterobacter* species.

Skin and Skin-Structure Infections, including those associated with post operative wounds, ulcers and burns caused by *Escherichia coli, Proteus mirabilis, Serratia marcescens, Enterobacter* species, *Pseudomonas aeruginosa, Klebsiella pneumoniae* and *Citrobacter* species*.

Intra-abdominal Infections, including peritonitis caused by *Escherichia coli, Klebsiella* species including *K. pneumoniae, Enterobacter* species including *E. cloacae**, *Pseudomonas aeruginosa, Citrobacter* species* including *C. freundii** and *Serratia* species* including *S. marcescens**.

Gynecologic Infections, including endometritis and pelvic cellulitis caused by *Escherichia coli, Klebsiella pneumoniae**, *Enterobacter* species* including *E. cloacae** and *Proteus mirabilis**.

*Efficacy for this organism in this organ system was studied in fewer than ten infections.

AZACTAM (aztreonam) is indicated for adjunctive therapy to surgery in the management of infections caused by susceptible organisms, including abscesses, infections complicating hollow viscus perforations, cutaneous infections and infections of serous surfaces. AZACTAM is effective against most of the commonly encountered gram-negative aerobic pathogens seen in general surgery.

Concurrent Therapy

Concurrent initial therapy with other antimicrobial agents and AZACTAM is recommended before the causative organism(s) is known in seriously ill patients who are also at risk of having an infection due to gram-positive aerobic pathogens. If anaerobic organisms are also suspected as etiologic agents, therapy should be initiated using an anti-anaerobic agent concurrently with AZACTAM (see DOSAGE AND ADMINISTRATION). Certain antibiotics (e.g., cefoxitin, imipenem) may induce high levels of beta-lactamase *in vitro* in some gram-negative aerobes such as *Enterobacter* and *Pseudomonas* species, resulting in antagonism to many beta-lactam antibiotics including aztreonam. These *in vitro* findings suggest that such beta-lactamase inducing antibiotics not be used concurrently with aztreonam. Following identification and susceptibility testing of the causative organism(s), appropriate antibiotic therapy should be continued.

CONTRAINDICATION

Aztreonam is contraindicated in patients with known allergy to this antibiotic.

WARNINGS

Careful inquiry should be made for a history of hypersensitivity reaction to any antibiotic or other drugs. Antibiotics should be given with caution to any patient who has had some form of allergy, particularly to drugs. It is recommended that patients who have had immediate hypersensitivity reactions (e.g., anaphylactic or urticarial) to penicillins and/or cephalosporins should be followed with special care. If an allergic reaction to aztreonam occurs, discontinue the drug and institute supportive treatment as appropriate (e.g., maintenance of ventilation, pressor amines, antihistamines, corticosteroids). Serious hypersensitivity reactions may require epinephrine and other emergency measures.

PRECAUTIONS

General

In patients with impaired hepatic or renal function, appropriate monitoring is recommended during therapy.

If an aminoglycoside is used concurrently with aztreonam, especially if high dosages of the former are used or if therapy is prolonged, renal function should be monitored because of the potential nephrotoxicity and ototoxicity of aminoglycoside antibiotics.

The use of antibiotics may promote the overgrowth of nonsusceptible organisms, including gram-positive organisms (*Staphylococcus aureus* and *Streptococcus faecalis*) and fungi. Should superinfection occur during therapy, appropriate measures should be taken.

Carcinogenesis, Mutagenesis, Impairment of Fertility

Carcinogenicity studies in animals have not been performed.

Genetic toxicology studies performed *in vivo* and *in vitro* with aztreonam in several standard laboratory models revealed no evidence of mutagenic potential at the chromosomal or gene level.

Two-generation reproduction studies in rats at daily doses up to 20 times the maximum recommended human dose, prior to and during gestation and lactation, revealed no evidence of impaired fertility. There was a slightly reduced survival rate during the lactation period in the offspring of rats that received the highest dosage, but not in offspring of rats that received five times the maximum recommended human dose.

Pregnancy

Pregnancy Category B

Aztreonam crosses the placenta and enters the fetal circulation.

Studies in pregnant rats and rabbits, with daily doses up to 15 and 5 times, respectively, the maximum recommended human dose, revealed no evidence of embryo- or fetotoxicity or teratogenicity. No drug induced changes were seen in any of the maternal, fetal, or neonatal parameters that were monitored in rats receiving 15 times the maximum recommended human dose of aztreonam during late gestation and lactation.

There are no adequate and well-controlled studies in pregnant women. Because animal reproduction studies are not always predictive of human response, aztreonam should be used during pregnancy only if clearly needed.

Nursing Mothers

Aztreonam is excreted in breast milk in concentrations that are less than 1 percent of concentrations determined in simultaneously obtained maternal serum; consideration should be given to temporary discontinuation of nursing and use of formula feedings.

Pediatric Use

Safety and effectiveness have not been established in infants and children.

ADVERSE REACTIONS

Local reactions such as phlebitis/thrombophlebitis following IV administration, and discomfort/swelling at the injection site following IM administration occurred at rates of approximately 1.9 percent and 2.4 percent, respectively.

Systemic reactions (considered to be related to therapy or of uncertain etiology) occurring at an incidence of 1 to 1.3 percent include diarrhea, nausea and/or vomiting, and rash. Reactions occurring at an incidence of less than 1 percent are listed within each body system in order of decreasing severity:

Hypersensitivity—anaphylaxis.

Hematologic—pancytopenia, neutropenia, thrombocytopenia, anemia, leukocytosis, thrombocytosis.

Gastrointestinal—abdominal cramps; rare cases of *C. difficile*-associated diarrhea or gastrointestinal bleeding have been reported.

Dermatologic—purpura, erythema multiforme, urticaria, exfoliative dermatitis, petechiae, pruritus, diaphoresis.

Cardiovascular—hypotension, transient ECG changes (ventricular bigeminy and PVC).

Respiratory—one patient experienced flushing, chest pain, and dyspnea.

Hepatobiliary—hepatitis, jaundice.

Nervous System—seizure, confusion, vertigo, paresthesia, insomnia, dizziness.

Musculoskeletal—muscular aches.

Special Senses—tinnitus, diplopia, mouth ulcer, altered taste, numb tongue, sneezing and nasal congestion, halitosis.

Other—vaginal candidiasis, vaginitis, breast tenderness.

Body as a Whole—weakness, headache, fever, malaise.

Adverse Laboratory Changes

Adverse laboratory changes without regard to drug relationship that were reported during clinical trials were:

Hepatic—elevations of AST (SGOT), ALT (SGPT), and alkaline phosphatase; signs or symptoms of hepatobiliary dysfunction occurred in less than 1 percent of recipients (see above).

Hemic—increases in prothrombin and partial thromboplastin times, eosinophilia, positive Coombs test.

Renal—increases in serum creatinine.

OVERDOSAGE

If necessary, aztreonam may be cleared from the serum by hemodialysis and/or peritoneal dialysis.

DOSAGE AND ADMINISTRATION

AZACTAM (aztreonam) For Injection may be administered intravenously or by intramuscular injection. Dosage and route of administration should be determined by susceptibility of the causative organisms, severity and site of infection, and the condition of the patient.

AZACTAM DOSAGE GUIDE (ADULTS)

Type of Infection	Dose*	Frequency (hours)
Urinary tract infection	500 mg or 1 g	8 or 12
Moderately severe systemic infections	1 g or 2 g	8 or 12
Severe systemic or life-threatening infections	2 g	6 or 8

*Maximum recommended dose is 8 g per day.

The intravenous route is recommended for patients requiring single doses greater than 1 g or those with bacterial septicemia, localized parenchymal abscess (e.g., intra-abdominal abscess), peritonitis or other severe systemic or life-threatening infections. Because of the serious nature of infections due to *Pseudomonas aeruginosa*, dosage of 2 g every six or eight

hours is recommended, at least upon initiation of therapy, in systemic infections caused by this organism.

The duration of therapy depends on the severity of infection. Generally, AZACTAM should be continued for at least 48 hours after the patient becomes asymptomatic or evidence of bacterial eradication has been obtained. Persistent infections may require treatment for several weeks. Doses smaller than those indicated should not be used.

Renal Impairment

Prolonged serum levels of aztreonam may occur in patients with transient or persistent renal insufficiency. Therefore, the dosage of AZACTAM should be halved in patients with estimated creatinine clearances between 10 and 30 mL/min/1.73 m² after an initial loading dose of 1 g or 2 g.

When only the serum creatinine concentration is available, the following formula (based on sex, weight, and age of the patient) may be used to approximate the creatinine clearance (Clcr). The serum creatinine should represent a steady state of renal function.

$$\text{Males: Clcr} = \frac{\text{weight (kg)} \times (140 - \text{age})}{72 \times \text{serum creatinine (mg/dL)}}$$

Females: 0.85 x above value

In patients with severe renal failure (creatinine clearance less than 10 mL/min/1.73 m²), such as those supported by hemodialysis, the usual dose of 500 mg, 1 g or 2 g should be given initially. The maintenance dose should be one-fourth of the usual initial dose given at the usual fixed interval of 6, 8 or 12 hours. For serious or life-threatening infections, in addition to the maintenance doses, one-eighth of the initial dose should be given after each hemodialysis session.

Dosage In The Elderly

Renal status is a major determinant of dosage in the elderly; these patients in particular may have diminished renal function. Serum creatinine may not be an accurate determinant of renal status. Therefore, as with all antibiotics eliminated by the kidneys, estimates of creatinine clearance should be obtained, and appropriate dosage modifications made if necessary.

Preparation Of Parenteral Solutions

General

Upon the addition of the diluent to the container, contents should be shaken **immediately and vigorously.** Constituted solutions are not for multiple-dose use; should the entire volume in the container not be used for a single-dose, the unused solution must be discarded.

Depending upon the concentration of aztreonam and diluent used, constituted AZACTAM (aztreonam) For Injection yields a colorless to light straw yellow solution which may develop a slight pink tint on standing (potency is not affected). Parenteral drug products should be inspected visually for particulate matter and discoloration whenever solution and container permit.

Admixtures With Other Antibiotics

Intravenous infusion solutions of AZACTAM (Aztreonam For Injection) prepared with Sodium Chloride Injection USP 0.9% or Dextrose Injection USP 5%, to which clindamycin phosphate, gentamicin sulfate, tobramycin sulfate, or cefazolin sodium have been added at concentrations usually used clinically, are stable for up to 48 hours at room temperature or seven days under refrigeration. Ampicillin sodium admixtures with aztreonam in Sodium Chloride Injection USP 0.9% are stable for 24 hours at room temperature and 48 hours under refrigeration; stability in Dextrose Injection USP 5% is two hours at room temperature and eight hours under refrigeration.

Aztreonam-cloxacillin sodium and aztreonam-vancomycin hydrochloride admixtures are stable in Dianeal® 137 (Peritoneal Dialysis Solution) with 4.25% Dextrose for up to 24 hours at room temperature.

Aztreonam is incompatible with nafcillin sodium, cephradine, and metronidazole.

Other admixtures are not recommended since compatibility data are not available.

Intravenous (IV) Solutions

For Bolus Injection: The contents of an AZACTAM (aztreonam) For Injection 15 mL capacity vial should be constituted with 6 to 10 mL Sterile Water for Injection USP.

For Infusion: Contents of the 100 mL capacity bottle should be constituted to a final concentration not exceeding 2% w/v (at least 50 mL of any appropriate infusion solution listed below per gram aztreonam). Most solutions may be frozen immediately after constitution in the original container (see Stability below).

If the contents of a 15 mL capacity vial are to be transferred to an appropriate infusion solution, each gram of aztreonam should be initially constituted with at least 3 mL Sterile Water for Injection USP. Further dilution may be obtained with one of the following intravenous infusion solutions:

- Sodium Chloride Injection USP, 0.9%
- Ringer's Injection USP
- Lactated Ringer's Injection USP
- Dextrose Injection USP, 5% or 10%
- Dextrose and Sodium Chloride Injection USP, (5%:0.9%), (5%:0.45%) or (5%:0.2%)
- Sodium Lactate Injection USP (M/6 Sodium Lactate)
- Ionosol® B and 5% Dextrose
- Isolyte® E
- Isolyte® E with 5% Dextrose
- Isolyte® M with 5% Dextrose
- Normosol®-R
- Normosol®-R and 5% Dextrose
- Normosol®-M and 5% Dextrose
- Mannitol Injection USP, 5% or 10%
- Lactated Ringer's and 5% Dextrose Injection
- Plasma-Lyte® M and 5% Dextrose
- 10% Travert® Injection
- 10% Travert® and Electrolyte No. 1 Injection
- 10% Travert® and Electrolyte No. 2 Injection
- 10% Travert® and Electrolyte No. 3 Injection

Intramuscular (IM) Solutions

The contents of an AZACTAM (aztreonam) For Injection 15 mL capacity vial should be constituted with at least 3 mL of an appropriate diluent per gram aztreonam. The following diluents may be used:

- Sterile Water for Injection USP
- Bacteriostatic Water for Injection USP (with benzyl alcohol or with methyl- and propylparabens)
- Sodium Chloride Injection USP, 0.9%
- Bacteriostatic Sodium Chloride Injection USP (with benzyl alcohol or with methyl- and propylparabens)

Stability Of IV And IM Solutions

AZACTAM (aztreonam) solutions for IV infusion at concentrations not exceeding 2% w/v must be used within 48 hours following constitution if kept at controlled room temperature (59° - 86°F/15° - 30°C) or within seven days if refrigerated (36° - 46°F/2° - 8°C).

Frozen aztreonam infusion solutions (except solutions prepared with the following two diluents, which have not been tested: Mannitol Injection 10%; Lactated Ringer's and 5% Dextrose Injection), may be stored for up to three months at -4° F/-20° C; frozen solutions may be thawed at controlled room temperature or by overnight refrigeration. Solutions that have been thawed and maintained at controlled room temperature or under refrigeration should be used within 24 or 72 hours after removal from the freezer, respectively. Solutions should not be refrozen.

AZACTAM solutions at concentrations exceeding 2% w/v, except those prepared with Sterile Water for Injection USP or Sodium Chloride Injection USP, should be used promptly after preparation; the two excepted solutions must be used within 48 hours if stored at controlled room temperature or within seven days if refrigerated.

Intravenous Administration

Bolus Injection: A bolus injection may be used to initiate therapy. The dose should be slowly injected directly into a vein, or the tubing of a suitable administration set, over a period of three to five minutes (see next paragraph regarding flushing of tubing).

Infusion: With any intermittent infusion of aztreonam and another drug with which it is not pharmaceutically compatible, the common delivery tube should be flushed before and after delivery of aztreonam with any appropriate infusion solution compatible with both drug solutions; the drugs should not be delivered simultaneously. Any AZACTAM infusion should be completed within a 20 to 60 minute period. With use of a *Y-type administration set*, careful attention should be given to the calculated volume of aztreonam solution required so that the entire dose will be infused. A *volume control administration set* may be used to deliver an initial dilution of AZACTAM (aztreonam) For Injection (see Preparation Of Parenteral Solutions, For Infusion) into a compatible infusion solution during administration; in this case, the final dilution of aztreonam should provide a concentration not exceeding 2% w/v.

Intramuscular Administration

The dose should be given by deep injection into a large muscle mass (such as the upper outer quadrant of the gluteus maximus or lateral part of the thigh). Aztreonam is well tolerated and should not be admixed with any local anesthetic agent.

HOW SUPPLIED

AZACTAM For Injection (Aztreonam For Injection)

Single-dose 15 mL capacity vials:
500 mg/vial: Packages of 10 (NDC 0003-2501-10) and 25 (NDC 0003-2501-15)
1 g/vial: Packages of 10 (NDC 0003-2502-10) and 25 (NDC 0003-2502-15)
2 g/vial: Packages of 10 (NDC 0003-2503-10) and 25 (NDC 0003-2503-15)

Single-dose 100 mL capacity intravenous infusion bottles with ball bands:
500 mg/bottle: Packages of 10 (NDC 0003-2501-20)
1 g/bottle: Packages of 10 (NDC 0003-2502-20)
2 g/bottle: Packages of 10 (NDC 0003-2503-20)

Storage

Store original packages at room temperature; avoid excessive heat.

E. R. Squibb & Sons, Inc.
Princeton, NJ 08540

J4-140

Issued December 1986

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DIV.
DIRECTOR
REVIEW

NDA 50-580
Aztreonam

December 30, 1986

Division Director's Memorandum

The following is an addendum to my memorandum of December 19, 1986, listed by numbers in that memorandum.

1. The addition of the word "rare" is acceptable.

2. The submitted further safety update information dated December 29, 1986 for meningitis, pediatric, CF, and neutropenia studies is acceptable.

E. Tabor

Edward Tabor, M.D.

cc:
Orig NDA
HFN-815
HFN-815/JLew
HFN-815/CSO
HFN-815/ETabor:mas-12/31/86-0403d

Orig NDA

NDA 50-580
Azactam

December 19, 1986

Division Director's Memorandum

The following issues still must be addressed:

1. The approval letter must include a revised "Gastrointestinal" subsection of ADVERSE REACTIONS as follows: "abdominal cramps; cases of C. difficile-associated diarrhea or gastrointestinal bleeding have been reported."

2. The cover letter of the October 28, 1986 Safety Update implies that it does not include the meningitis, pediatric, CF, and neutropenia studies. This must be determined.

3. Microbiology. They have just added "multiply" by the letter of November 6, 1986. This can be left for the time being, since it is used in other drug labels.

4. The gonorrhea claim could have been given for women on the basis of the 87 U.S. females and 103 U.K. females. (Of the 87 U.S. females, clinical outcome was measured in only 61; the bacteriologic cure in the 87 was 98.9%. Of the 103 U.K. females, clinical outcome was measured only in 45 (25 symptomatic and 20 asymptomatic) but the bacteriologic cure rate in the 103 was 97%.)

   A discussion was held in the Division, and it was felt that this meets our requirement for 100 females with a greater than 95% cure rate. The use of foreign cases to make up the 100 is not significant, since a relatively large number (87) were U.S. females. The issue of symptoms in females is not a major concern, since so many females with this infection are asymptomatic and have to be treated.

   However, for men, only 107/115 evaluable patients (94%) were cured. In fact, in the Handsfield Study, only 23/28 (82%) were cured. These results are unacceptable for a gonorrhea indication.

5. The P.I.D. claim cannot be given without the gonorrhea claim. Even if the gonorrhea claim were given, the P.I.D. claim could not be given because the protocol was not followed with regard to follow-up culture. Only 10 evaluable patients remain.

6. In MICROBIOLOGY, first paragraph, last sentence: delete "and clinical infections." There are several on the list that are not in the INDICATIONS.

   Alternatively, organisms which do not belong on the first list could be moved to the second list.

   Both lists of microorganisms must have our usual statement re: only microorganisms listed in the INDICATIONS have been shown clinically, etc. At present, the first list has no statement and the second list has an inadequate statement.

7. Asterisk statement should be placed at the end of the list of systems in the INDICATIONS.

8. INDICATIONS

   Gyn - delete N. gonorrhoeae.
   Uncomplicated gonorrhea - delete it.

9. WARNINGS

   Delete "Preliminary evidence indicates that patients with documented penicillin allergy do not react to aztreonam; however".

10. Carcinogenesis etc.

    The correct heading should be used.

11. Use: "Pregnancy: Pregnancy category B".

12. Adverse Reactions:

    a) omit first sentence;

    b) change second sentence to include percentage of patients with local reactions; omit "were more frequent than systemic reactions in clinical studies."

These points were conveyed to the company in two phone calls, one from Ms. Creedon and one from myself, on December 19, 1986.

E. Tabor

Edward Tabor, M.D.

cc:
Orig NDA
HFN-815
HFN-815/MO
HFN-815/CSO
HFN-815/ETabor:mas-12/19/86-0391d

MED REVIEW

Addendum to Medical Officer's Review of NDA 50-580
(Printed Package Insert)

Review Begun: December 19, 1986
Review Completed: December 22, 1986

Sponsor: E.R. Squibb & Sons
New Brunswick, New Jersey 08903

Drug Name: Trade: AZACTAM
generic: aztreonam

Submitted December 18, 1986 are the protocols concerning treatment of N. gonorrhoeae pelvic infections as requested in the November, 1986 review.

Reason for Submission:
To get approval for use of aztreonam in pelvic gynecologic infections due to N. gonorrhoeae.

Review:
Study Design: Protocols: 18554-73/18554-16A: These were randomized, comparative studies comparing an aztreonam regimen to a standard treatment regimen in the treatment of serious gram-negative infections. For the purpose of this review, only N. gonorrhoeae pelvic infections are evaluated.

Treatment period ranged from 4 to 7 days.

Patient Population:
Females 18 years or older with post-obstetric/gynecologic infections (with no excluding factors present).

Documentation of diagnosis was by clinical signs and symptoms of pelvic soft tissue infection and by a positive pretherapy culture for a susceptible N. gonorrhoeae within 48 hours before starting therapy. Isolation of the organism was required from endocervical fluid, peritoneal fluid, biopsy specimen or blood culture to make the diagnosis. All the patients cases in this review had positive pretherapy endocervical cultures.

Efficacy Evaluation:
1. Repeat culture after 2 to 5 days and at the end of therapy if specimen available.
2. Salpingitis due to N. gonorrhoeae must have repeat endocervical cultures within 4 to 7 days post-therapy to document cure.
3. Investigator is assessment of clinical response with clinical evaluation 7 to 14 days post-therapy.

Safety data was not provided and its evaluation will not be addressed in this MOR.

Drug Dosage:
Aztreonam 1 gram or 2 grams TID.

Protocol 18554-41: Significantly deviated from the above study design in that a follow-up culture 4-7 days post-treatment was not required for proof of cure. The single case report submitted for review under this protocol was unevaluable because no susceptibility testing to the test drug aztreonam was done.

Investigators:
Protocol 18554-16A; (17 evaluable* patients)
Dr. Melvin Dodson
Protocol 18554-73; (12 evaluable* patients)
Dr. Subin Roy
Protocol 18554-41; (1 evaluable* patient)
Dr. Richard Sweet

*evaluable according to the sponsor. Significantly less patients were found evaluable in this MOR.

Results:

Table 1

Total Patient No.	30
Evaluable Patients	8
Reasons unevaluable:	
Improper follow-up cultures*:	20
Susceptibility Not Done:	2

*19/20 patients, contrary to protocol, had their follow-up culture on the last day of therapy. One patient had her follow-up culture 2 days post-therapy vs. 4-7 days.

Table 2

Evaluable Patients	8
Race:	
Caucasian	4
Black	4
Age Range	19-29 years
Dosage:	
1 gram TID	4 patients
2 gram TID	4 patients

Table 3

Response:	Bacteriologic[1]	Clinical[2]
Success:	8	8
Failure:	0	0

1. Bacteriologic success in this MOR is defined as negative cultures for N. Gonorrhoeae at the appropriate follow-up intervals (4-7 days post-therapy).

2. Clinical success in this MOR is defined as defervescence and complete resolution of signs and symptoms of the infection under study.

Conclusions:

Not enough evaluable patients were studied to allow the claim that azactam is effective for N. gonorrhoeae pelvic infections, although all 8 evaluable patients (out of 30 submitted for review) had both bacteriological and clinical cures.

Also note that 2 different dosage regimens were used in the 8 cured evaluable patients. With so few patients evaluable, it is impossible to assess if both regimens are equally as effective. For future reference, note: The Azactam Dosage Guide in the package insert (p. 13 of 17 of draft) may allow for a potentially less efficacious dosage regimen than that used in the studies on GC pelvic infections. In these studies, 1 or 2 grams of Azactam was used TID, whereas if such infections are considered moderately severe infections, a potential regimen of 1 gram BID is recommended in the insert.

Recommendations:

The claim for efficacy in N. gonorrhoeae pelvic infections be denied.

Judy F. Lew, M.D.

cc:
Orig
HFN-340
HFN-815 ST 12/29/86
HFN-815/CSO
HFN-815/JFLew/llm/12/23/86
1877m

JRD 29Dec86

12/16/86

Medical Officer's Review of NDA 50-580
(Printed Package Insert)

Date of Submission: December 10, 1986
Date Received by MO: December 12, 1986
Review Completed: December 15, 1986

Sponsor: E.R. Squibb & Son
New Brunswick, New Jersey 08903

Drug Name: Trade: AZACTAM
generic: aztreonam

Reason for Submission: To seek approval for final printed package insert.

Review/Conclusions:

The sections: Description, Clinical Pharmacology, Microbiology, Susceptibility Testing are to be also reviewed by the appropriate person(s) in the Pharmacology or Microbiology divisions.

Note: A prior review of NDA 50-580 submitted November 13, 1986 and completed November 24, 1986 addressed changes in the given package insert labeling concerning:

1. indications of aztreonam for N. Gonorrhoeae infection as on page 9 of package insert draft/page 3 of final package insert,

2. use of the word "multiply-resistant" on page 5 of draft/page 2 of final,

3. a statement suggesting aztreonam may not cause C. difficile - associated diarrhea or abnormal bleeding (page 12 of draft/page 3 of final),

4. evidence of synergism between aztreonam and aminoglycosides (page 6 of draft/page 2 of final).

Please refer to the above review for the respective conclusions and recommendations.

Note:

On page 12 of draft/page 3 of final under "Adverse Reactions", "(Ventricular bigeminy and PVC)" has been added to "Cardiovascular" reactions. The incident(s) that prompted this inclusion is not clear, but since virtually any adverse reaction is possible secondary to drugs, no object is raised in its inclusion under potential reactions.

Note:

Listed under "Warnings" (page 10 of draft and page 3 of final):

"Preliminary evidence indicates that patients with documented penicillin allergy do not react to aztreonam; however,"....

Exactly what evidence supports this claim and if the evidence is adequate to justify the above statement is not clear. The suggestion that there is no cross-reactivity may be premature if based on a small and/or inadequate study.

Note:

Except for the pertinent items mentioned above, the final package insert submitted appears to have incorporated prior FDA recommended revisions.

Recommendations:

1. Please refer to the appropriate reviews by pharmacology and microbiology.

2. Recommendations as in the November 1986 review.

3. The statement "Preliminary evidence indicates that patients with documented penicillin allergy do not react to aztreonam," be eliminated or revised pending submission of further evidence justifying the implied claim.

Suggestion for Revision:

There is some evidence that patients with documented penicillin allergy may not react to aztreonam ....

4. The information that prompted the inclusion of ventricular bigeminy and PVC as a possible adverse effect of aztreonam be submitted.

Judy Lew, M.D.

cc:
Orig NDA
HFN-340
HFN-815 ET 1/15/87
HFN-815/CSO
HFN-815/JLew:bam:12/16/86:1858m

ORD 29Dec86

Medical Officer's Review on NDA 50-580
(Amendment)

Date of Submission: November 13, 1986
Received by MO: November 18, 1986
Date Completed: November 24, 1986

Sponsor: E. R. Squibb & Sons
New Brunswick, NJ 08903

Drug Name: Trade: Azactam
Generic: aztreonam

Reason for Submission:

1. To obtain approval for aztreonam use as a single 1 gram intramuscular dose for uncomplicated gonorrhea in women.

2. To obtain approval for aztreonam use for pelvic gynecologic infection due to N. gonorrhoeae.

3. To obtain approval for a revised draft insert reflecting FDA's "approvable" labeling for aztreonam as communicated in the FDA letter of October 16, 1986 plus the following additions:

   a. inclusion of N. gonorrhoeae under Gynecologic Infections (indications)
   b. similarly, the inclusion of uncomplicated gonorrhea caused by penicillinase and non-penicillinase - producing strains as an indication.
   c. inclusion of the word "multiply" in the context of:

      Aztreonam ... is effective in laboratory animal models and clinical infections against most strains of the following organisms, including many that are multiply-resistant to other antibiotics (i.e., certain cephalosporins, penicillins, and aminoglycosides):

      Escherichia coli
      Enterobacter species

      etc.

   d. inclusion of a sentence on synergistic activity in vitro with aminoglycosides in the microbiology section of the insert.

Review on aztreonam for uncomplicated gonorrhea in women:

1. Submitted are an additional 10 women studied under protocol 18554-10c previously filed with the FDA in December, 1983. Earlier, 79 females with uncomplicated gonorrhea infection who received aztreonam were found to be evaluable in the previous MOR. In concurrence with FDA standards, more evaluable female patients were required before aztreonam could be approved for use in such infections.

II. Also submitted is narrative summary of a multicentered study done in the United Kingdom evaluating the use of aztreonam IM in the treatment of acute uncomplicated gonorrheal infection. (Protocol AB. AZT. 002)

I. Study Design of Protocol 18554-10
Briefly: Patients with presumptive diagnosis of gonorrhea infection were entered into the study; confirmation was by culture isolation of N. gonorrhoeae. Patients were randomly assigned to receive a single 1 gram dose of aztreonam or a 2 gram dose of spectinomycin IM. Follow-up clinical and bacteriological examinations were done between 3 and 7 days after completion of the single dose therapy.

Submitted in this supplement are 10 female patients who received aztreonam as their IM medication.

Investigators:

Gary Slutkin, M.D.
San Francisco, CA

Thomas W. Austin, M.D.
Ontario, Canada

Hugh Robson, M.D.
Montreal, Quebec, Canada

Results:

No. of Patients:	10				
Reasons unevaluable:					
Sensitive pathogen not obtained	1				
Follow-up cultures not done	1				
Evaluable patients:	8	Success		Failure	
Infection site:					
Cervix only	3	3			
Cervix + rectum	3	2		1	
Cervix + throat	1	1			
rectum only	1	1			
		7/8	87%	1/8	13%

Combined Results[1]:

Evaluable Patients:	87	Success[2]		Failure	
Infection site:					
Cervic (C) only	21	21			
Rectum (R) only	2	2			
C + R	4	3	75%	1	25%
C + urethra	51	51			
C + throat	1	1			
C + R + throat	1	1			
C + R + urethra	7	7			
Total		86/87	98.9%	1/87	1.1%

1. Includes results of patients found evaluable by Dr. Min in her MOR of the December, 1983 NDA.

2. Success is bacteriologic only; 26 patients were not evaluable for the clinical response in the prior MOR.

Penicillinase - producing strains of N. gonorrhoeae were isolated in 23 women in the aztreonam group and 24 women in the spectinomycin group. All of these women had microbiologic cures on their respective therapies.

II. Study Design of Protocol AB. AZT. 002

This was an open uncontrolled multicenter study done in the United Kingdom, aztreonam given as a single 1 gram IM dose for patients with uncomplicated genital and/or anorectal gonorrhoeae. Confirmation of the infection was made by culture and sensitivity to aztreonam determined.

At entry, clinical signs and symptoms were recorded. Between 2 and 21 days after aztreonam administration, patients returned for repeat cultures to determine microbiological cure.

Exclusions:

1. Age under 18 or over 75 years.
2. History of anaphylactic reaction or other serious reaction to penicillins or cephalosporins.
3. Pregnancy or breast feeding.
4. Presence of a condition requiring an antiinfective agent other than the study drug.
5. Neutrophil count less than 1,000 per mm^3 (if done).
6. Any other condition which, in the opinion of the investigator would make the patient unsuitable for enrollment.
7. History of antibiotics, including metronidazole, in the prior 14 days or during therapy and follow-up (excluding antifungal agents).

Results of female patients only were reported in this supplement.

Investigators:

Dr. Lester Cohen
Cardiff, UK

Dr. John R. W. Harris
London, UK

Dr. Richard A. Sparks
Birmingham, UK

Dr. Robert C. Spencer
Sheffield, UK

Dr. Ivan B. Tait
Glasgow, UK

Demographic data was not submitted beyond the ages of the patients, which ranged from 16 to 53 years in the 168 women who were given aztreonam.

Results:*

Number of patients enrolled:	168				
Reasons unevaluable:					
No N. gonorrhoeae isolated:	11				
Susceptibility not recorded	24				
Lost to follow-up	10				
Follow-up day not given	13				
Concurrent antibiotic	7				
Evaluable patients	103				
Site of infection:	Total	Success**		Failure	
Urethra (U)	1	1			
Cervix (C)	10	10			
U + C	62	60	(96.8%)	2	(3.2%)
C + Pharynx (P	1	1			
U + C + Rectum	22	22			
P + U + C + R	7	6	(85.7%)	1	(14.3%)
Total	103	100/103	(97.1%)	3/103	(2.9%)

*Results are as determined by the sponsor.

**Microbiological cure only. For patients with multiple sites, microbiologic cures were claimed only when all sites were cured. Clinical response was unevaluable by the sponsor in 78 women; 20 were asymptomatic at enrollment and 58 had concomitant GU infection with other pathogens that may have masked clinical response.

Five out of eight women with penicillinase - producing strains of N. gonorrhoeae were evaluable according to the sponsor. All five of these women had microbiologic cures following aztreonam administration.

Safety and Tolerance:
Nine of 168 patients treated with aztreonam experienced 11 adverse events possibly due to aztreonam. All complaints according to the sponsor resolved spontaneously without the need for specific treatment.

Adverse Effect	Patient No.
1. Injection site pain	1
2. Leg discomfort	3
3. Injection site mass	1
4. Vomiting	2
5. Loose stools	1
6. Abdominal pain	1
7. Abdominal rash	1
8. Headache	1

Conclusion on aztreonam for treatment of uncomplicated gonorrhea:
Although only 87 female patients were evaluable in the comparative studies done on the efficacy of aztreonam in treatment of uncomplicated GC infections, the efficacy was adequate with a 98.9% cure rate, including a 100% cure rate in 23 patients with penicillinase producing N. gonorrhoeae.

Also, there was 97.1% cure rate in 103 female patients in an uncontrolled, open study that included cures in all 5 patients who had penicillinase producing N. gonorrhoeae. Unfortunately, little demographic information is provided to enable a better review of this study.

Despite this shortcoming, these studies combined suggest that aztreonam use should be effective for uncomplicated gonorrhea infections with penicillinase and non-penicillinase producing strains.

Pelvic Gynecologic Infection Due to N. Gonorrhoeae
The patient summary data was pooled from 3 different studies under protocols: 18,554-16A, 18,554-41 and 18,554-73 Addendum D. Since descriptions of the protocols were not submitted in this NDA supplement, the review will be held pending acquisition of protocol information that would enable an adequate review.

Package Insert Labeling Proposal:
In accordance with FDA's "approvable" labeling for aztreonam, as communicated in October, 1986, the aztreonam label was changed.

Comments on other label proposals:

1. The word "multiply-resistant" in the context of aztreonam being effective against: many [strains of organisms] that are "resistant" to other antibiotics ...seems unnecessary and may be confusing. The definition of "multiply-resistant" is unclear. "Multiply" as an adjective suggests "especially" or possibly "many more times"; either definition seems inappropriate.

2. Data suggesting that aztreonam and aminoglycosides have been shown to be synergistic against certain strains of P. aeruginosa, Enterobacter cloacae and other gram-negative aerobic bacilli was not submitted in this NDA supplement.

3. The statement, "Rare cases of C. difficile-associated diarrhea or abnormal bleeding have been reported in patients who were previously or concomitantly treated with other drugs including antibiotics or chemotherapeutic agents; the relationship to aztreonam is unclear.", may be misleading. The incidence of C. difficile-associated G.I. adverse reactions reported during use with aztreonam is the same as that with many other broad spectrum antibiotics, around 0.05 to 0.15%. Until there is better clinical justification, this statement suggesting that aztreonam may not cause pseudomembranous colitis could be misleading.

Recommendations:

1. Aztreonam use as a single 1 gram intramuscular dose for uncomplicated N. gonorrhoeae (penicillinase and non-penicillinase producing) infections in women be approved.

2. Approval for aztreonam use for pelvic gynecologic infection due to N. gonorrhoeae be held until the appropriate protocol description information is (again) submitted to enable completion of the associated review.*

3. The word "multiply-resistant" be changed back to "resistant" as in the original label proposal.

4. The statement suggesting that aztreonam may not cause C. difficile-associated diarrhea or abnormal bleeding be revised to reflect the similar reported incidence of this adverse reaction as compared to many other broad spectrum antibiotics (i.e., certain cephalosporins).

5. Data suggesting that aztreonam and aminoglycosides are synergistic be (again) submitted for review before the associated label change be allowed.

*Note: It is acknowledged that the data requested has been submitted before and reviewed by a different medical officer who is presently no longer available at FDA to make the appropriate recommendations pertaining to the above proposals.

Judy F. Lew, M.D.

cc:
Orig NDA
HFN-340
HFN-815
HFN-815/CSO
HFN-815/JFLew/llm/11/25/86
1796m

12/19/86 Division Director's Comment
See Division Director's Memorandum
S.T.

DRO 12Dec86

Medical Officer's Review of Safety Update Report for NDA 50-580

Date of Submission: October 31, 1986
Received by MO: November 4, 1986
Date Completed: November 14, 1986

Sponsor: E. R. Squibb & Sons
New Brunswick, NJ 08903

Drug name: Trade: Azactam
generic: aztreonam

Reason of submission:
The document submitted additional safety data generated between November, 1985 and October, 1986 as requested by FDA in accordance with regulation 314.50(d) (5) (vi) (b).

A tabulation of adverse reactions comparing the NDA data base, the November, 1985, initial safety update data base and the current information generated between November, 1985 and October, 1986 are presented and reviewed in this MOR.

Review:

The following data is the same as that submitted by the company. A review of the case reports did not produce any significant discrepancies.

1. Single dose studies: No change in status since November, 1985.

2. Multidose studies: A post-update (November, 1985 to October, 1986) patient population (N=353) is compared to the NDA data base (N=1771) and to the November, 1985 data base (N=4570). The incidence of clinical adverse drug reactions (ADRs) was 10.8% in the October update vs. 6.9% in the NDA and 6.5% in the November update. The incidence of discontinuations due to ADRs was 3.1% vs. 1.6% and 1.5% respectively and 0.3% vs. 0.5% and 0.4% respectively for the laboratory abnormalities.

The reported incidence of death was 1.1% in the October update vs. 0.5% in the NDA and 0.7% in the November update.

See attached charts page 2, 3 and 4.

NDA 50-580

Chart 2

Table I

CLINICAL ADVERSE REACTIONS (ADRs)
MULTIDOSE STUDIES

	NDA (N = 1771)		NOV 1985 UPDATE (N = 4570)		POST-UPDATE (N = 353)	
REACTION	NUMBER OF ADRS	% OF PATIENTS	NUMBER OF ADRS	% OF PATIENTS	NUMBER OF ADRS	% OF PATIENTS
Dermatologic	29	1.6%	82	1.8%	13	3.7%
Rash	18	1.0%	59	1.3%	9	2.5%
Rash with Eosinophilia	6	0.3%	11	0.2%	1	0.3%
Pruritus	2	0.1%	8	0.2%	3	0.8%
Purpura	3	0.2%	-	0.09%	-	-
Gastrointestinal	39	2.2%	105	2.3%	7	2.0%
Diarrhea	13	0.7%	47	1.0%	4	1.1%
Nausea/Vomiting	16	0.9%	26	0.6%	1	0.3%
Taste Alteration	4	0.2%	10	0.2%	1	0.3%
Colitis/*C. difficile* diarrhea	2	0.1%	13	0.3%	-	-
Jaundice/"Hepatitis"	3	0.2%	5	0.1%	-	-
Oral Lesions	1	0.06%	4	0.09%	1	0.3%
Local Reactions•	43	2.4%	82	1.8%	16	4.5%
Phlebitis/Thrombophlebitis	33	2.3%*	64	1.7%**	15	4.6%♦
Discomfort/Swelling at Injection Site	10	2.8%***	18	2.3%****	1	3.3%♦♦
CNS-Related	7	0.4%	24	0.5%	2	0.6%
Headache	3	0.2%	11	0.2%	1	0.3%
Dizziness	2	0.06%	5	0.1%	1	0.3%
Other	2	0.06%†	8	0.2%††		
Miscellaneous	12	0.7%	45	1.0%	6	1.7%
Drug Fever	0	-	3	0.07%	2	0.6%
Pyrexia/Chills/Cold Sweats	1	0.06%	8	0.2%	1	0.3%
Vaginitis	2	0.1%	7	0.2%	1	0.3%
Fatigue	2	0.1%	5	0.1%	-	-
Hypersensitivity Reaction	1	0.06%	2	0.04%	-	-
Bleeding	2	0.1%	4	0.09%	-	-
Other	4	0.2%†††	16	0.4%††††	2•••	0.6%
Total: Adverse Reactions	130		338		44	
Total: Patients	123	6.9%	299	6.5%	38	10.8%

*Based on the 1410 patients who received aztreonam intravenously.
**Based on the 3804 patients who received aztreonam intravenously.
♦Based on the 323 patients who received aztreonam intravenously.
***Based on the 361 patients who received aztreonam intramuscularly.
****Based on the 766 patients who received aztreonam intramuscularly.
♦♦Based on the 30 patients who received aztreonam intramuscularly.
†One case of each of the following: confusion, seizure.
††One case of each of the following: disturbed mental processes, impaired hearing in one ear, seizure and insomnia. Two cases of vertigo were reported in addition to †)
†††One case of each of the following: subclavian vein thrombosis, bloating and swelling, abdominal cramps and hypotension
††††One case of each of the following: paresthesia, flushing, dyspnea, chest pain, lower limb edema, bradycardia, abdominal pain, diplopia, coughing, sweating, warmness in feet and ECG abnormality (in addition to †††).
•Includes all patients specially reported as having had an "Adverse Reaction." It does not include data collected under case report page designed for assessments of tolerance.
•••Kidney pain (1), muscular twitching and clonic motion (1).

NDA 50-580

Chart 3:

Appendix 2

DISCONTINUED PATIENTS
CLINICAL ADVERSE REACTIONS
MULTIDOSE STUDIES

REACTION	NDA (N = 1771)		NOV 1985 UPDATE (N = 4570)		POST-UPDATE (N = 353)	
	NUMBER OF ADRS	% OF PATIENTS	NUMBER OF ADRS	# OF PATIENTS	NUMBER OF ADRS	# OF PATIENTS
Dermatologic						
Rash	9	0.5%	30	0.7%	7	(2%)
Rash with Eosinophilia	6	0.3%	9	0.2%	-	-
Purpura	2	0.1%	2	0.04%	-	-
Gastrointestinal						
Diarrhea	2	0.1%	3	0.1%	1	(0.3%)
Nausea/Vomiting	1	0.06%	2	0.04%	-	-
Taste Alteration	-		2	0.04%	-	-
Colitis/C. *difficile* diarrhea	-		4	0.1%	-	-
Jaundice/"Hepatitis"	1	0.06%	2	0.04%	-	-
Local Reactions						
Phlebitis/Thrombophlebitis	3	0.2%	3	0.1%	1	(0.3%)
CNS-Related						
Headache	-		1	0.02%	-	-
Other*	2	0.1%	3	0.1%	-	-
Miscellaneous						
Drug Fever	-		1	0.02%	2	(0.6%)
Pyrexia/Chills/Cold Sweats	-		5	0.1%	-	-
Fatigue	-		1	0.02%	-	-
Hypersensitivity Reaction	1	0.06%	2	0.04%	-	-
Bleeding	2	0.1%	2	0.04%	-	-
Other**	1	0.06%	6	0.2%	-	-
Total: Adverse Reactions	30		78		11	
Total: Patients	29	1.6%	68	1.5%	11	(3.1%)

NDA *Confusion
Seizure

**Bloating and Swelling

UPDATE *Confusion
Seizure
Vertigo

**Bloating and Swelling
Coughing
Flushing
Dyspnea
Chest Pain
Diplopia

NDA 50-580

Chart 4

Appendix 5

CAUSES OF DEATH
ALL PROTOCOLS

CAUSES OF DEATH	NDA (N = 2117)	NOV 1985 UPDATE (N = 5013)	POST-UPDATE (N = 353)
Cardiac Arrhythmia/Arrest	15 (0.7%)	43 (0.8%)	3 (0.8%)
Myocardial Infarct	11 (0.5%)	13 (0.3%)	4 (1.1%)
Heart Failure	11 (0.5%)	20 (0.4%)	1 (0.3%)
Bleeding and Shock	5 (0.2%)	11 (0.2%)	- -
Other Cardiovascular Causes (Hypotension)	1 (<0.1%)	4 (<0.1%)	1 (0.3%)
Pulmonary Embolism	6 (0.3%)	17 (0.3%)	2 (0.6%)
Respiratory Failure	5 (0.2%)	26 (0.5%)	4 (1.1%)
Other Respiratory Causes (Aspiration Pneumonia)	3 (0.1%)	6 (0.1%)	- -
Cerebrovascular Accident	2 (<0.1%)	12 (0.2%)	1 (0.3%)
Renal Failure	1 (<0.1%)	4 (<0.1%)	1 (0.3%)
Multiple Organ Failure	12 (0.6%)	20 (0.4%)	2 (0.6%)
Overwhelming Infection	34 (1.6%)	107 (2.1%)	6 (1.7%)
Malignancy	13 (0.6%)	31 (0.6%)	3 (0.8%)
Operative/Postoperative/ Posttraumatic Complications	1 (<0.1%)	11 (0.2%)	- -
Miscellaneous	10* (0.5%)	33** (0.7%)	4*** (1.1%)

*[Massive intestinal ischemia (1), Perforation stomach (1), Sudden death (1), Unknown (1), and Not stated (6)]

**[Duodenal perforation (1), Hepatic failure (2), Burns (1), Hypoglycemia (2), Suicide (1), Car accident (1), Collagen Vascular Disease (2), Electrolyte disturbance (1), Sudden death (1), Refusal of blood transfusion Aspiration of gastric contents (1), Not stated (9)]. These are in addition to cases reported in *.

***Cystic fibrosis (2), complications of rheumatoid arthritis (1), Meningitis (1)

Conclusions:
The higher incidence of clinical adverse reactions in the October, 1986 update, 10.8%, vs. prior reported incidences of 6.9 and 6.5% reflects mostly increase of rash and phelbitis/thrombophlebitis reported. The incidence of rash (2.5%) and phelbitis/thrombophlebitis (4.6%) in the October 1986 update are approximately twice the incidence reported before. The significance of this is unclear; it could reflect better reporting, chance, or that the population upon which the studies were performed have characteristics predisposing them to more adverse reactions, i.e., cystic fibrosis patients, many who have allergic characteristics and who require long term therapy. In view of the significantly smaller data base in the October 1986 update compared to prior reports, no definite change in the incidence of adverse reactions can be made.

Judy F. Lew, M.D.

cc:
Orig NDA
HFN-340
HFN-815 97 12/19/86
HFN-815/RNorton
HFN-815/CSO
HFN-815/JFLew/llm/11/20/86
1777m

DRD 1 dec 86

March 26, 1986

NDA 50-580

16.1

Group Leader's Follow-up Comments

Drug Name: Aztreonam

As noted in my earlier comments on Dr. Min's review of the aztreonam NDA, she used throughout her review criteria for efficacy that often are not those customarily used by other reviewers in the Division.

Because of this, the sponsor (Squibb Laboratories) felt that they were being unfairly treated and were being denied claims that would have been approved if the usual Divisional standards of efficacy had been used.

I therefore volunteered to review the efficacy results for all patients reviewed by Dr. Min, if the company would provide the results when efficacy was judged by the standards customarily used in the Division. I agreed to discuss my conclusions, and all discrepancies, with Dr. Min and to arbitrate all differences.

I have finished my review of the data provided by Squibb, and have today discussed in detail my conclusions with Dr. Min.

Evaluation: My assessments of efficacy based on the material supplied by Squibb resulted in my granting fewer claims than were granted by Dr. Min. There were several errors in her review where claims that should have been obviously granted based on the conclusions in her review were not included in her final summary. These inadvertent exclusions have been corrected. We discussed in detail the borderline areas where she had granted claims, and I had not. They were claims where small numbers of organisms had been studied and where Dr. Min felt that her conclusions based on her detailed review of the data in the NDA justified the claims being granted, and I therefore defer to her judgement.

Conclusions:

Although some of Dr. Min's assessments of efficacy were more stringent than those customarily used by other members of the Division, the end result was that she granted more claims based on her review of the data than I would have granted from a summary of data where efficacy had been assessed by customary criteria. The sponsor therefore doe not have a basis for claiming that they have been unjustly denied valid claims.

Recommendations:

Form 5, 50-580, should be found approvable for the indications specified in Dr. Min's review dated November 12, 1985.

Labeling can be agreed upon while the Form 5 is under review in HFN-800.

George R. Stanley, M.D.
Group Leader, DAIDP

cc:
Form 5, 50-580
HFN-340
HFN-815 GT 4/2/86
HFN-815/CSO
HFN-815/GRStanley
0380m

Form 5 50-580

Addendum to MOR of Form 5 50-580 (Azactam for Injection) dated 11/12/85

March 4, 1986

The following tables may provide the additional information which was sought by the supervisors in their reviews (12/26/85 and 1/27/86) of the 11/12/85 MOR, regarding the controlled clinical studies of urinary tract infections (UTIs) and lower respiratory tract infections (LRTIs).

Protocol 18554-11 Comparison of Aztreonam and Tobramycin in the Treatment of Aerobic Gram-negative Lower Respiratory Tract Infections.

Microbiologic Response by Investigators

Investigator Number	Number cured*/Number treated			
	Aztreonam		Tobramycin	
	Applicant**	MO	Applicant**	MO
6228***	27/28 (96%)	23/24 (96%)	12/12 (100%)	11/11 (100%)
6449***	19/20 (95%)	14/20 (70%)	1/5 (25%)	1/5 (25%)
7614***	9/15 (60%)	7/16 (44%)	2/5 (40%)	2/6 (33%)
6226***	8/8 (100%)	8/8 (100%)	4/4	4/4
6345****	8/8 (100%)	8/9 (89%)	2/2	1/2
6227***	3/6 (50%)	3/6 (50%)	0/1	0/1
6317***	3/3	3/3	1/1	1/1
6366*****	2/2	2/2	1/1	1/1
3096***	1/1	1/2	0/1	0/1
6207***	1/1	1/1	1/2	0/1
6224****	1/2	1/2	-	-
6229****	1/1	1/1	-	-
6401****	0/1	0/1	-	-

*Microbiological cure was assumed where clinical improvement together with absence of sputum production was seen during and/or after completion of therapy.
**Applicant's analysis (from a table in Vol. 3.3; p 11).
MO - Medical Officer's analysis.
****US investigator
*****Foreign investigator.

Protocol 18554-11 Comparison of Aztreonam and Tobramycin in the Treatment of Aerobic Gram-negative Lower Respiratory Tract Infections.

Pathogen	Number eradicated*/Number treated**		
	Aztreonam	Tobramycin	Moxalactam
P. aeruginosa	16/27 (59%)	4/12 (33%)	-
E. coli	17/18 (94%)	3/3	-
K. pneumoniae	15/16 (94%)	6/7 (86%)	1/1
H. influenzae	11/12 (92%)	3/3	3/3
Enterobacter aerogenes	4/5	1/1	-
E. cloacae	4/5	4/4	-
Enterobacter sp.	2/2	1/1	-
Klebsiella oxytoca	3/4	1/1	1/1
Proteus mirabilis	4/5	4/8	-
P. vulgaris	1/1	-	-
Serratia sp.	2/2	1/1	-
S. marcescens	1/1	0/1	-
S. rubidaea	1/1	-	-
Providencia stuartii	1/1	-	-
Morganella morganii	1/1	-	-
Citrobacter diversus	1/1	1/1	-
H. parainfluenzae	1/1	-	1/1***
Acinetobacter sp.	-	1/1	-
Total	85/103 (82.5%)	30/44 (68.2%)	6/6

*Microbiological eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.
** Pooled from domestic and foreign studies; Number include isolates from single and multiple-pathogen infections.
***This patient received both tobramycin and moxalactam.

Aztreonam in the Treatment of Serious Gram-negative Urinary Tract Infections*

Pathogen	Microbiologic Response** Number eradicated/Number treated Uncomplicated UTI 5-9 d	Uncomplicated UTI 4-6 wk	Complicated UTI 5-9 d	Complicated UTI 4-6 wk	Total 5-9d	Total 4-6 wk
E. coli	63/71 (89%)	51/68 (75%)	40/54 (74%)	32/51 (63%)	103/125 (82%)	83/119 (70%)
K. pneumoniae	8/8 (100%)	6/7 (86%)	16/18 (89%)	13/17 (76%)	24/26 (92%)	19/24 (79%)
P. mirabilis	8/8 (100%)	7/8 (88%)	8/9 (89%)	7/9 (78%)	16/17 (94%)	14/17 (82%)
P. aeruginosa	5/5	4/5	9/16 (56%)	5/15 (33%)	14/21 (67%)	9/20 (45%)
E. cloacae	4/4	3/4	5/5	4/5	9/9 (100%)	7/9 (78%)
E. aerogenes	0/1	0/1	0/1	0/1	0/2	0/2
K. oxytoca	1/1	1/1	3/3	3/3	4/4	4/4
P. stuartii	1/1	1/1	0/1	0/1	1/2	1/2
P. fluorescens	1/1	1/1	-	-	1/1	1/1
C. diversus	1/1	1/1	1/1	1/1	2/2	2/2
Citrobacter sp.	-	-	2/2	1/2	2/2	1/2
C. freundii	-	-	2/2	1/2	2/2	1/2
S. marcescens	1/1	1/1	3/3	3/3	4/4	4/4
Serratia sp.	-	-	1/1	1/1	1/1	1/1
P. vulgaris	-	-	2/2	2/2	2/2	2/2
P. rettgeri	-	-	2/2	2/2	2/2	2/2
M. morganii	-	-	1/1	1/1	1/1	1/1
Total	93/102 (91.2%)	76/98 (77.6%)	95/121 (78.5%)	76/116 (65.5%)	188/223 (84.3%)	152/214 (71.0%))

*Data pooled from the controlled studies (protocols 18554-13, 14, 27 and 28).
**At 5-9 days and 4-6 weeks afer completion of therapy.

Treatment of Serious Gram-negative Urinary Tract Infections*

Pathogen	Microbiologic Response** Number eradicated/Number treated Aztreonam 5-9 d	Aztreonam 4-6 wk	Cefamandole 5-9 d	Cefamandole 4-6 wk	Aminoglycoside 5-9d	Aminoglycoside 4-6 wk
E. coli	103/125 (89%)	83/119 (75%)	41/55 (75%)	27/47 (57%)	11/13 (85%)	7/11 (64%)
K. pneumoniae	24/26 (92%)	19/24 (79%)	6/9 (67%)	6/8 (63%)	- -	- -
P. mirabilis	16/17 (94%)	14/17 (82%)	3/6 (50%)	2/5 (40%)	- -	- -
P. aeruginosa	14/21 (67%)	9/20 (45%)	- -	- -	4/4	3/3
E. cloacae	9/9 (100%)	7/9 (78%)	2/2	2/2	-	-
E. aerogenes	0/2	0/2	2/2	2/2	-	-
K. oxytoca	4/4	4/4	1/1	-	1/1	1/1
P. stuartii	1/2	1/2	0/1	0/1	-	-
P. fluorescens	1/1	1/1	-	-	-	-
C. diversus	2/2	2/2	0/1	0/1	-	-
Citrobacter sp.	2/2	1/2	-	-	-	-
C. freundii	2/2	1/2	1/1	1/1	-	-
S. marcescens	4/4	4/4	-	-	-	-
Serratia sp.	1/1	1/1	-	-	-	-
P. vulgaris	2/2	2/2	2/2	1/1	-	-
P. rettgeri	2/2	2/2	-	-	-	-
M. morganii	1/1	1/1	-	-	1/1	1/1
Total	188/223 (84.3%)	152/214 (71.0%)	58/80 (72.5%)	41/68 (60.3%)	17/19 (89.5%)	12/16 (75.0%)

*Data pooled from the controlled studies (protocols 18554-13, 14, 27 and 28).
**At 5-9 days and 4-6 weeks afer completion of therapy.

Aztreonam in the Treatment of Serious Gram-negative Urinary Tract Infections*

Pathogen	Microbiologic Response** Number eradicated/Number treated Aztreonam 5-9 d	4-6 wk
P. aeruginosa	22/28 (78.6%)	20/26 (76.9%)
E. coli	5/5	4/5
K. pneumoniae	1/2	1/2
E. cloacae	4/4	4/4
E. aerogenes	2/2	2/2
K. oxytoca	4/4	4/4
P. rettgeri	5/5	5/5
P. stuartii	2/2	2/2
C. freundii	1/1	1/1
M. morganii	2/2	2/2
Total	48/55 (87.3%)	45/53 (84.9%)

*Data from the uncontrolled study (protocols 18554-31).
**At 5-9 days and 4-6 weeks afer completion of therapy.

Aztreonam and Amoxicillin in the Treatment of Uncomplicated Lower Urinary tract Infectons caused by Aerobic Gram-negative Organisms (Protocol 18554-15)*

	Microbiologic Response** Number eradicated/Number treated			
	Aztreonam		Amoxicillin	
Pathogen	5-9 d	4-6 wk	5-9 d	4-6 wk
E. coli	41/50 (82%)	38/50 (76%)	36/37 (97%)	31/35 (89%)
K. pneumoniae	3/4	1/1	1/1	1/1
E. aerogenes	1/1	1/1	-	-
Citrobacter sp.	1/1	1/1	-	-
P. mirabilis	-	-	2/2	2/2
Total	46/56 (82.1%)	41/53 (77.4%)	39/40 (97.5%)	34/38 (89.5%)

*Aztreonam (single I.M. dose) therapy vs. amoxicillin (conventional multiple oral dose) therapy.
*At 5-9 days and 4-6 weeks afer completion of therapy.

Pathogen	Microbiologic Response Number eradicated/Number treated Urinary Tract Infections (UTIs) C(a) 5-9 d	UC(b) 5-9d	C(a) 4-6 wk	UC(b) 4-6 wk	LRTI C(c)	UC(d)
E. coli	103/125 (89%)	-	83/119 (75%)	35/50 (70%)	17/18 (94%)	7/9 (78%)
K. pneumoniae	24/26 (92%)	-	19/24 (79%)	13/15 (87%)	15/16 (94%)	12/15 (80%)
P. mirabilis	16/17 (94%)	-	14/17 (82%)	3/5	4/5	6/8 (75%)
P. aeruginosa	14/21 (67%)	-	9/20 (45%)	27/53 (51%)	16/27 (59%)	14/46 (30%)
E. cloacae	9/9 (100%)	-	7/9 (78%)	5/7 (71%)	4/5	3/4
E. aerogenes	0/2	-	0/2	2/2	4/5	5/6 (83%)
Enterobacter sp.	-	-	-	1/1	2/2	1/1
E. hafniae	-	-	-	-	-	1/1
K. oxytoca	4/4	-	4/4	5/5	3/4	1/1
P. fluorescens	1/1	-	1/1	-	-	-
Citrobacter sp.	2/2	-	1/2	-	-	-
C. diversus	2/2	-	2/2	1/1	1/1	2/2
C. freundii	2/2	-	1/2	1/1	-	2/2
Serratia sp.	1/1	-	1/1	1/1	2/2	2/2
S. marcescens	4/4	-	4/4	2/4	1/1	4/5
S. rubidaea	-	-	-	-	1/1	-
P. vulgaris	2/2	-	2/2	-	1/1	-
P. rettgeri	2/2		2/2	6/6	-	-
P. stuartii	1/2	-	1/2	3/4	1/1	-
M. morganii	1/1	-	1/1	3/3	1/1	-
Providencia sp.	-	-	-	0/1	-	-
Haemophilus sp.	-	-	-	-	-	1/1
H. influenzae	-	-	-	-	11/12 (92%)	22/24 (92%)
H. parainfluenzae	-	-	-	-	1/1	-
Klebsiella sp.	-	-	-	1/1	-	-
Pseudomonas sp.	-	-	-	0/1	-	1/6
Total	188/223 (84.3%)	-	152/214 (71.0%)	109/161 (67.7%)	85/103 (82.5%)	84/133 (63.2%)

Note:

UTIs (urinary tract infections):
C(a) - Data pooled from the controlled studies (protocols 18554-13, 14, 27 & 28)
UC(b) - Data pooled from the uncontrolled (noncomparative)studies (Protocols 18554-31 &-16).

LRTIs (lower respiratory tract infections):
C(c) - Data from the multicenter controlled study (protocol 18554-11)
UC(d) - Data from the uncontrolled (noncomparative)study (protocols 18554-16)

F. Min, M.D.

Orig Form 5 50-580
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HFN-815/CSO
HFN-340
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HFN-815/Norton
HFN-815/Min:fm/3/4/86
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ERD 31 March 86

10.1

Group Leader's Comments on MOR for Form 5 50-580 dated February 21, 1986

March 4, 1986

Applicant: E. R. Squibb and Sons

Name of Drug: generic: aztreonam
Trade: Azactam for injection

Comments: I have read Dr. Min's review and concur with most of her comments and conclusions.

However, I take exception with her conclusions about the adequacy of the bone and joint infection studies. I agree with her denying the claim, but feel that she has done so for the wrong reason. Dr. Min has turned down th claim on the basis that the sponsor has not done a comparative study of the treatment of bone and joint infections. It has not been Division policy tc require comparative studies for all claims granted. We have routinely insisted that two major claims, one of them being systemic, must be proven by comparative studies. Other claims could be granted based on open studies, provided there were not over-riding public health concerns (ie. gonorrhea, meningitis).

We have not required comparative studies as essential for granting a claim in osteomyelitis, and even if we were to decide now that they are needed, it is inappropriate to use the new standard for studies that were started in good faith some four years ago using the previously accepted standard.

Conclusion:

The sponsor has studied so few cases for acute and chronic osteomyelitis and and for acute septic arthritis that I feel it is appropriate to turn down the claim. However, it is not appropriate to require comparative studies to approve a claim for use in bone and joint infections.

George R. Stanley

George R. Stanley, M.D.
Group Leader, DAIDP

cc:

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Form 5 50-580

9.1

Medical Officer's Review of Form 5 50-580 Amendments

February 21, 1986

Applicant: E. R. Squibb and Sons

Name of Drug: Trade - Azactam for Injection
Generic - Aztreonam

Category of Drug: Monobactam antibiotic

Date of Form 5 Amendments: November 13, 1985 (vol. 8.1 - 8.4) and December 16, 1985 (vol. 9.1 - 9.2).

Reasons for Amendments: Additional clinical data to support proposed claims.

Medical Officer's Review of Original Form 5 Submission: Refer to MOR dated November 12, 1985.

Background: In response to our earlier request, the applicant has submitted additional clinical data on controlled studies of intra-abdominal and obstetric/gynecologic infections, and open studies of bone and joint infections to augment the limited clinical data on these indications accumulated since the initial Form 5 submission. Published clinical reports by a few investigators who were participants to the open or comparative studies of aztreonam were also submitted. This was intended to update and augument the limited clinical data on these indications.

Evaluation and Comments:

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus Clindamycin in the Treatment of Intra-abdominal Infections

This was a multicenter, randomized, comparative study of aztreonam vs. tobramycin in the treatment of intra-abdominal infections. The amendment included additional clinical data provided by 5 principal investigators, 2 domestic and 3 foreign. As shown in Table I (A), the two domestic investigators (#5099 and #6407) treated 9 patients, 4 in the aztreonam group and 5 in the tobramycin group. Investigator #5099 treated one patient each with the test and the control drugs. Since the post-therapy follow-up in the tobramycin-treated patients was short (2 days after completion of therapy), this case as well as the aztreonam-treated patient were excluded from the efficacy analyses by this reviewer. One foreign investigator (#6376) treated only one patient with aztreonam; this case was also excluded from the efficacy analyses. Two foreign investigators (#6444 and #7612) had 69 evaluable patients, 34 in the aztreonam group and 35 in the tobramycin group. The demographic characteristics of the two treatment groups were similar, as shown in Tables I (A) and II (A), and these were similar to those of earlier cases reviewed in that the patient population in the foreign studies were younger than the domestic patient population. The majority of patients were diagnosed as having peritonitis secondary to ruptured viscus or intra-abdominal abscess.

In the domestic studies, all patients received the drug intravenously, but in the foreign studies quite a few patients received the drugs both intravenously and intaramuscularly, or intramuscularly alone. All patients were treated concurrently with clindamycin. The number of patients who underwent surgery was comparable in the two treatment groups. The duration of patient follow-up in the domestic and foreign studies ranged from 6 to 32 days. The mean duration of follow-up was comparable in the two treatment groups.

The microbiological and clinical responses seen in the domestic and foreign studies are presented in Tables I (B) and II (B), respectively. Admittedly, the total number of additional domestic cases treated was small, but the favorable response to the drug therapy which was suggested in the earlier domestic cases reviewed is also noted in the additional cases. In the foreign studies, bacteriological cure was seen in 34 (100%) of the 34 aztreonam-treated patients and in 32 (91%) of the 35 tobramycin-treated patients. Clinical cure (resolution of symptoms and signs compatible with intra-abdominal infection) was seen in 30 (88.2%) of the aztreonam group and 32 (91.4%) of the tobramycin group. The overall microbiologic response of clinical isolates to the drug therapy is shown in Table III. The microbiological and clinical responses in the two treatment groups were similar. In the domestic studies, superinfection occurred in one tobramycin-treated patient but in none of the aztreonam-treated patients. In the foreign studies, superinfection occurred in 4 (11.8%) of the 35 patients in the aztreonam group and 3 (8.6%) of the 35 patients in the tobramycin group. The superinfection was due to gram-positive organisms in the aztreonam-treated patients, and gram-positive and gram-negative organisms in the tobramycin-treated patient (Tables I B and II B). Clinical failure was due to superinfection in these patients.

The microbiological and clinical outcome by investigator was as follows:

Investigator Number	Number Cured*/Number Treated AZT + CLI Micr	AZT + CLI Clin	TOB + CLI Micr	TOB + CLI Clin
Domestic:				
6407	3/3	3/3	2/3	2/3
Foreign:				
6444	19/19 (100%)	16/19 (84.2%)	22/24 (91.7%)	21/24 (87.5%)
7612	15/15 (100%)	14/15 (93.3%)	10/11 (90.9%)	11/11 (100%)
Total	37/37 (100%)	33/37 (89.2%)	34/38 (89.5%)	34/38 (89.5%)

* The criteria for the microbiological (Micr) and clinical (Clin) cures were based on the resolution of symptoms and signs consistent with the diagnoses during and at post-therapy (6 days or longer after completion of therapy) and/or the eradication of the initial pathogen(s).

The safety of the drugs was assessed in 81 patients who were treated with the test or the control drug.
Adverse reactions which were possibly or probably related to drug therapy were observed in 23 (58%) of the 40 patients in the aztreonam group and in 33 (80%) of the 41 patients in the tobramycin group. This incidence of adverse reactions in the additional cases was arbitrarily high because the applicant did not provide data on the non-evaluable patients who had received the drugs. The significance of the difference between the two treatment groups, therefore, could not be ascertained.

The most frequent adverse reactions were mild to moderate local reactions at the infusion or injection site. Laboratory abnormalities noted were primarily transient elevation of hepatic enzymes (aminotransferases/alkaline phosphatase) and eosinophilia. The laboratory abnormalities were not accompanied by a clinical manifestation of hepatic dysfunction or allergy. A few patients in each treatment group had more than one adverse reaction. In none of the patients was the drug discontinued because of the adverse reactions.

The reactions observed in the domestic and foreign studies were as follows:

Number of Patients Treated	AZT/CLI 40	TOB/CLI 41
Clinical:		
Diarrhea	19	26
Nausea	1	3
Local Reactions at infusion of injection site(thrombophlebitis; pain, erythema,and/or induration)	0	1
	18	25
Laboratory abnormalities:		
Eosinophilia	13	16
Increased AST(SGOT)/ALT(SGPT)	6	2
Increased alkaline phosphatase	2	6
Increased LDH	2	2
Thrombocytopenia	0	1
Thrombocytosis	0	1
Casts/protein in urine	3	2
	0	2

AZT - Aztreonam TOB - Tobramycin CLI - Clindamycin

Deaths occurred in 3 patients, 2 in the aztreonam group, and 1 in the tobramycin group, during and/or after therapy. The deaths were not attributable to the drugs.

Conclusions: The results for the additional patients entered into this multicenter, randomized study indicated that aztreonam, as an adjunct to surgery, is as effective and safe as the aminoglycoside, tobramycin, in the treatment of intra-abdominal infections caused by aerobic gram-negative pathogens, when these drugs were concomitantly used with an effective anti-anaerobic drug, clindamycin. The results of the additional cases evaluated lessen this reviewer's earlier concern about the small number of patients studied. The earlier results of the non-comparative studies of aztreonam in the treatment of intra-abdominal infections were also supportive of the favorable results seen in the multicenter comparative study. Approval of the indication intra-abdominal infections caused by susceptible aerobic gram-negative organisms (E. coli, Enterobacter species, including E. cloacae, Klebsiella pneumoniae, Pseudomonas aeruginosa, Serratia species* including S. marcescens, and Citrobacter species* including C. freundii) is therefore recommended.(see Table IV).

Note: * signifies that the organisms were the pathogen(s) in less than 10 (but more than 5) evaluable cases.

Table I (A)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Aerobic Gram-negative Intra-abdominal Infections (Domestic Study)

Number and (ID No.) of Principal Investigators: 2 (5099*; 6407)

	Treatment AZT + CLI	TOB + CLI
Number of Patients Evaluable for Efficacy:		
by Applicant	4	6
by MO	3	3
Reasons for Exclusion:		
Inadequate or no post-therapy follow-up	0	2
Other (No evaluable patients in the control group)	1	0
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	2	2
Male	1	1
Age (years)		
Range	47 - 88	76 - 83
Mean	71.7	79.3
Race		
Caucasian	1	3
Other (not stated)	2	0
Diagnoses:		
Peritonitis (ruptured viscus)	3	3
Dosage Regimen:(IV):	2 g q 8-12 h	80-100 mg q 8-12h
Duration of Treatment (days):		
Range	8 - 10	5 - 16
Mean	8.7	9.0
Surgery during Therapy	2	2

*The investigator did not enter evaluable patients into both treatment groups.
MO - Medical officer
AZT - aztreonam
TOB - tobramycin
C.I - clindamycin

Table I (B)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Aerobic Gram-negative Intra-abdominal Infections (Domestic Study)

Microbiological Response*

Gram-negative Pathogens	No. Eradicated/No. Treated AZT + CLI	TOB + CLI
Single Pathogen:		
Escherichia coli	-	0/1
Klebsiella pneumoniae	-	1/1
Pseudomonas aeruginosa	-	1/1
Citrobacter freundii	1/1	--
Multiple Pathogens:		
E. coli + P. aeruginosa	1/1	-
E. coli + C. freundii + K. oxytoca	1/1	-
Total	3/3	2/3
Superinfection: E. aerogenes	0/3 -	1/3 1

*The eradication of microorganisms was assumed when the resolution of symptoms and signs consistent with the diagnoses occurred during and at post-therapy(6 days or longer after completion of therapy) and/or the follow-up cultures were negative for the initial pathogen(s).

AZT - aztreonam
CLI - clindamycin
TOB - tobramycin

Table II (A)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Aerobic Gram-negative Intra-abdominal Infections (Foreign Study)

No. of Principal Investigators & Investigators' Number: 3 (6376*; 6444, 7612)

	Treatment AZT + CLI	TOB + CLI
Number of Patients Evaluable for Efficacy:		
by Applicant	37	36
by MO	34	35
Reasons for Exclusion:		
Inadequate or no post-therapy follow-up	1	0
Other (No evaluable patients in the control group; other diagnosis)	2	1
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	14	9
Male	20	26
Age (years)		
Range	14 - 81	10 - 76
Mean	36.2	31.3
Race		
Caucasian	24	30
Black	7	3
Other (or not stated)	3	2
Diagnosis		
Peritonitis (appendicitis/ruptured viscus/abscess)	34	35
Dosage Regimen:	1 g q 6-8h	50 - 75 mg q 8 h
Route of Administration		
IV	18	13
IV & IM	9	10
IM	4	11
Not stated	3	1
Duration of Therapy (Days)		
Range	5 - 15	5 - 14
Mean	8.4	9.2
Surgery during Therapy	4	4

*The investigator did not enter the evaluable patients into both treatment groups.

MO - Medical officer AZT - aztreonam TOB - tobramycin CLI - clindamycin

Table II (B)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Aerobic Gram-negative Intra-abdominal Infections (Foreign Study)

Microbiological Response*

Gram-negative Pathogen	AZT + CLI Number eradicated/	TOB + CLI No. Treated
Single Pathogen:		
E. coli	16/16	18/18
Enterobacter sp.	3/3	1/1
Klebsiella pneumoniae	3/3	3/3
Klebsiella sp.	-	2/2
K. oxytoca	1/1	-
P. aeruginosa	2/2	1/2
Proteus mirabilis	-	1/1
P. rettgeri	-	1/1
P. vulgaris	-	3/3
Proteus sp.	1/1	-
Citrobacter diversus	1/1	-
Multiple pathogens:		
E. coli + Enterobacter sp.	1/1	-
E. coli + K. pneumoniae**	2/2	0/1
E. coli + Klebsiella sp.	1/1	-
E. coli + Proteus sp.	1/1	-
E. coli + P. aeruginosa**	-	1/2
Enterobacter sp. + Klebsiella sp.	1/1	
Enterobacter sp. + K. oxytoca	-	1/1
E. coli + Enterobacter sp + K. pneumoniae	1/1	-
Total	34/34 (100.0%)	32/35 (91.4%)
Superinfection:	4/34 (11.8%)	3/35 (8.6%)
Staphylococcus aureus	2	1
S. aureus + Streptococcus sp./ S. faecalis	2	0
S. aureus + P. aeruginosa	0	1
E. coli	0	1

*The eradication of microorganisms was assumed when the resolution of symptoms and signs consistent with the diagnoses occurred during and at post-therapy(6 days or longer after completion of therapy) and/or the follow-up cultures were negative for the initial pathogen(s).
AZT - aztreonam CLI - clindamycin TOB - tobramycin

Table III

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Aerobic Gram-negative Intra-abdominal Infections (Domestic and Foreign Studies)

Microbiologic Response*

Gram-negative Pathogen	AZT + CLI	TOB + CLI
	Number of Isolates eradicated/No. Treated	
E. coli	24/24 (100%)	22/24 (92%)
Enterobacter sp.	6/6	2/2
Klebsiella pneumoniae	6/6	5/6
Klebsiella sp.	2/2	2/2
K. oxytoca	2/2	1/1
P. aeruginosa	3/3	4/6
Proteus sp.	2/2	-
P. vulgaris	-	3/3
P. mirabilis	-	1/1
Providencia rettgeri	-	1/1
Citrobacter diversus	1/1	-
C. freundii	2/2	-
Total	48/48 (100%)	41/46 (89%)

*The eradication of microorganisms was assumed when the resolution of symptoms and signs consistent with the diagnoses occurred during and at post-therapy(6 days or longer after completion of therapy) and/or the follow-up cultures were negative for the initial patnogen(s).

AZT - aztreonam
CLI - clindamycin
TOB - tobramycin

Table IV

Aztreonam plus Clindamycin in the Treatment of Aerobic Gram-negative Intra-abdominal Infections (Domestic and Foreign Studies)*

Microbiological Response**

Gram-negative Pathogen	Controlled studies Number of Isolates eradicated/ No. Treated	Uncontrolled Studies Number of Isolates eradicated/ No. Treated
E. coli	35/35 (100%)	27/30 (90%)
Enterobacter sp./E. cloacae	8/9	3/4
Klebsiella pneumoniae	8/9	12/12 (100%)
Klebsiella sp.	2/2	0/1
K. oxytoca	2/2	2/2
P. aeruginosa	6/6	12/16 (75%)
Proteus sp.	2/2	-
P. vulgaris	1/1	-
P. mirabilis	1/1	1/1
Citrobacter diversus/C. species	1/1	1/1
C. freundii	2/3	2/2
Serratia liquefaciens	1/1	1/1
S. marcescens	-	3/4
Aeromonas hydrophila	-	1/1
Aeromonas sp.	-	1/1
Pseudomonas sp.		1/1
Total***	69/72 (95.8%)	67/77 (87.0%)

*This represents pooled data on the earlier cases and the additional cases reviewed.
**The eradication of microorganisms was assumed when the resolution of symptoms and signs consistent with the diagnoses occurred during and at post-therapy (6 days or longer after completion of therapy) and/or the follow-up cultures were negative for the initial pathogen(s).
*** The total number of isolates treated was larger since a few patients had polymicrobial infections.

Clinical Response

Controlled studies Number cured or improved / No. Treated	Uncontrolled Studies Number cured or improved / No. Treated
47/53 (88.7%)	52/56 (92.9%)

Protocol 18554-41 : Comparison of Aztreonam plus Clindamycin with Gentamicin plus Clindamycin in the Treatment of Aerobic Gram-negative Obstetric and Gynecologic Infections

Domestic Study

This was a multicenter, randomized study, in which the efficacy and safety of aztreonam was compared to that of gentamicin, an aminoglycoside, in the treatment of obstetric and gynecologic infections.

The applicant's additional data consists of the computer case summary sheets and microfiches for the 66 cases which were considered by the applicant as evaluable for efficacy. These cases were provided by 6 principal investigators, 5 domestic and 1 foreign. Investigator #6435 enrolled the majority of the patients (36), as in the earlier submission. One domestic investigator (#4886) failed to follow the clinical protocol and did not randomize patients. His data, therefore, were analysed separately as an uncontrolled study, as shown in Tables IV and V. One other domestic investigator (#7535) did not enroll evaluable patients in the aztreonam treatment group. Only one foreign investigator (#6470) entered one patient each into the two treatment groups; the patients, however, had urinary tract infections, rather than gynecologic infections. The entrance criteria, clinical and bacteriological monitoring of the patients, and treatment regimens of the test and control drugs did not differ from those in the earlier studies. The demographic characteristics of the patients in the two treatment groups were comparable with regard to age and race, as presented in Table I. Endomyometritis (post-C section) was the predominant clinical diagnosis in the two treatment groups. The majority of patients had mixed aerobic and anaerobic infections. Clindamycin was concomitantly administered to all patients. The therapeutic results were analysed by this reviewer in patients who had the post-therapy follow-up not less than 7 days after completion of therapy, and those with no post-therapy follow-up were excluded from the efficacy evaluation. The number of the evaluable patients therefore is smaller than that of the applicant.
The bacteriological and clinical responses seen in the test and the control groups were similar. The cure rates were 95% (18/19) in the aztreonam group and 91% (20/22) in the gentamicin group, as noted in Table II. The cure rate for the additional cases in the aztreonam group was comparable to that seen in the smaller number of the earlier cases. The incidence of superinfections (or colonizations) was similar in the two treatment groups, occurring in 2 (11%) of the aztreonam-treated patients and in 2 (10%) of the gentamicin-treated patients. The causative microorganisms were S. faecalis in the aztreonam group and S. faecalis, E. cloacae and P. aeruginosa in the gentamicin group. No other antibiotic therapy was given to these patients. The cure rates seen in the individual investigator's studies were similar, as shown in Table III.

The bacteriologic and clinical responses seen in the non-randomized study are presented in Table V. As in the randomized study, the majority of the patients had mixed aerobic and anaerobic infections, and E. coli was the predominant gram-negative organisms isolated. In all 13 patients therapeutic success was attained.

The safety of the two treatments was assessed only in the patients whose line summary and microfiche were provided by the applicant. Adverse effects, possibly or probably related to drug therapy, were observed in 8% (3/36) of the aztreonam-treated patients and in 11% (3/28) of the gentamicin-treated patients. The adverse reactions were thrombophlebitis in the aztreonam-treated patients, and impaired renal function (abnormal creatinine and/or BUN levels) in the gentamicin-treated patients. One of the gentamicin-treated patients also developed a "rash".

Conclusions: The additional data submitted by the applicant has increased somewhat the number of evaluable patients with aerobic gram-negative gynecologic infections. The total number of the patients evaluated in the controlled study remains small, but this drug appears to be as effective as the control drug, an aminoglycoside (gentamicin), in the treatment of OB/GYN infections caused by Gram-negative organisms, particularly E. coli. Concurrent use of antianaerobic agent was necessary since a majority of the patients had mixed aerobic and anaerobic infections. The additional cases studied in an uncontrolled (non-comparative) study supported the findings of the controlled study. Approval of the indication OB/Gyn infections caused by aerobic gram-negative pathogens (E. coli, Proteus mirabilis*, Klebsiella pneumoniae*, and Enterobacter species*) is recommended (see Table VI).

Note: * signifies that the organisms were the pathogen(s) in less than 10 (but more than 5) evaluable cases.

Table I

Protocol 18554-41 : Comparison of Aztreonam plus Clindamycin with Gentamicin plus Clindamycin in the Treatment of Aerobic Gram-negative Obstetric and Gynecologic Infections (Domestic Study)

Number and (ID No.) of Principal Investigators: 4 (5178, 6435, 7535*, 7653)

	Treatment	
	AZT + CLI	GEN + CLI
Number of Patients Evaluable for Efficacy:	19	22
Demography and Other Characteristics of Evaluable Pts:		
Age (years)		
Range	16 - 35	18 - 36
Mean	21.0	25.2
Race		
Caucasian	15	15
Black	4	6
Not stated	0	1
Clinical Diagnosis:		
Endomyometritis	14	15
PID	4	5
Cellulitis(vaginal cuff)	0	1
Post-surgical wound infection	1	0
Polymicrobial (anaerobic/aerobic gram positive) infection	13/19 (68%)	19/22 (86%)
Dosage Regimen:	1 - 2 g q 8 h	1 - 1.5 mg/kg q 8 h (plus clindamycin 600 mg q 6 h I.V.)
Route of Administration:		
I.V.	15	16
I.V./I.M.	4	6
Duration of Treatment (days):		
Range	4 - 7	4 - 12
Mean	5.4	6.2
Surgery during therapy	1	1

*The investigator did not enter evaluable patients into both treatment groups.
AZT - aztreonam GEN - gentamicin CLI - clindamycin
PID - pelvic inflammatory disease

Table II

Protocol 18554-41 : Comparison of Aztreonam plus Clindamycin with Gentamicin plus Clindamycin in the Treatment of Aerobic Gram-negative Obstetric and Gynecologic Infections (Domestic Study)

Microbiological/Clinical Response*

	No. Cured /No. of Pts. Treated AZT + CLI	GEN + CLI
Single Pathogen:		
E. coli	8/9	12/13
K. pneumoniae	2/2	2/2
E. aerogenes	1/1	-
P. mirabilis	1/1	-
P. morganii	1/1	-
E. agglomerans	-	1/1
Multiple Pathogens:		
E. coli + P. mirabilis	2/2	-
E. coli + K. pneumoniae	1/1	2/2
E. aerogenes + P. mirabilis	1/1	-
E. coli + E. cloacae + P. mirabilis	1/1	-
E. coli + H. influenzae	-	1/1
E. coli + P. aeruginosa	-	1/1
E. coli + K. pneumoniae + P. mirabilis	-	1/1
K. pneumoniae + P. morganii	-	0/1
Total	18/19 (94.7%)	20/22 (90.9%)

Table III

Investigator's ID Number	Bacteriological and Clinical Cure* No. Cured /No. of Patients Treated AZT/CLI	GEN/CLI
5178	1/1	1/1
6435	13/14 (92.9%)	13/15 (86.7%)
7653	4/4	6/6

* The criteria for bacteriologic and clinical cures were based on resolution of symptoms and signs consistent with infections/and or the eradication of pathogen during and at post-therapy follow-up.
AZT - aztreonam GEN - gentamicin CLI - clindamycin

Table IV

Aztreonam plus Clindamycin in the Treatment of Aerobic Gram-negative Obstetric and Gynecologic Infections (Domestic Study)

Number and (ID No.) of Principal Investigator: 1 (4889)*

	Treatment AZT + CLI
Number of Patients Evaluable for Efficacy:	13
Demography and Other Characteristics of Evaluable Patients:	
Age (years)	
Range	18 - 41
Mean	32
Race	
Caucasian	0
Black	13
Clinical Diagnosis:	
Endomyometritis/endometritis	4
Vaginal cuff cellulitis	8
PID (TOA)	1
Dosage Regimen:	Aztreonam 1 - 2 g. q 8 h, I.V. plus clindamycin 600 mg q 6 h, I.V.
Duration of Treatment (days):	
Range	4 - 9
Mean	5.5
Surgery during therapy	1

* This investigator was a participant in the Protocol 18554-41 controlled study, but he did not randomized his patients.

AZT - aztreonam
CLI - clindamycin
PID - pelvic inflammatory disease
TOA - tubo-ovarian abscess

Table V

Aztreonam plus Clindamycin in the Treatment of Aerobic Gram-negative Obstetric and Gynecologic Infections (Domestic Study)

Principal Investigator Number: 4889*

Microbiological/Clinical Response

Pathogen	No. Cured*/No. of Patients Treated AZT + CLI
Single Pathogen:	
E. coli	7/7
K. pneumoniae	2/2
N. gonorrhoeae	1/1
P. mirabilis	1/1
Multiple Pathogens:	
E. coli + K. pneumoniae	1/1
C. freundii + K. pneumoniae	1/1
Total	13/13 (100%)

* The criteria for the microbiological and clinical cure were based on resolution of symptoms and signs consistent with the infections during and at post-therapy (7 days or longer after completion of therapy) and/or the eradication of the initial pathogen(s).

AZT - aztreonam
GEN - gentamicin
CLI - clindamycin

Table VI

Aztreonam plus Clindamycin in the Treatment of Aerobic Gram-negative Obstetric and gynecologic Infections (Domestic Studies)*

Microbiological Response

Gram-negative Pathogen	Controlled studies Number of Isolates eradicated**	Uncontrolled Studies /No. Treated
E. coli	14/15 (93%)	7/8(88%)
Enterobacter sp.* (E. cloacae/E. aerogenes)	5/5	1/1
Klebsiella pneumoniae*	6/6	-
Proteus mirabilis*	5/5	1/1
P. aeruginosa	1/1	-
P. morganii	1/1	-
N. gonorrhoeae	-	1/1
Total***	32/33 (96.9%)	10/11 (90.9%)

*This represents pooled data on the earlier cases and the additional cases reviewed (protocols 18554-41 and -16).
**The eradication of microorganism(s) was assumed when resolution of symptoms and signs consistent with infections occurred during and at post-therapy and/or the follow-up cultures were negative for the initial pathogen(s).
*** The total number of isolates treated was larger since a few patients had polymicrobial infections.

Clinical Response

Controlled studies Number cured or improved	Uncontrolled Studies / No. Treated
26/27 (96.3%)	22/23 (95.7%)

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections due to Aerobic Gram-negative Organisms

Bone and Joint Infections:

Eleven patients were added by the applicant to its January 1985 database of the bone and joint infections. In addition, the applicant provided additional information concerning the duration of further follow-up of patients (13) included in the earlier review. The eleven patients, 7 domestic and 4 foreign, were entered into this open study by the 4 domestic and 3 foreign investigators. The dosage of aztreonam (2 g q 6-8 h) was the same as that used in the earlier patients, but in 2 foreign patients a lower dosage (1 g q 8-12 h) was used. In all but one foreign patient the drug was administered intravenously. The duration of therapy ranged from 4 to 8 weeks.

Analysis of the additional 11 cases is presented in Table I. As in the cases reviewed earlier, the post-therapy follow-up period was rather short. It ranged from 10 days to 2 months. The favorable microbiologic and clinical responses observed in these patients, therefore, could not be ascertained as microbiologic and clinical cure, particularly in chronic osteomyelitis cases. Analysis of all evaluable patients (earlier cases plus additional cases) in whom the post-therapy follow-up was considered adequate (the follow-up period not less than one month for both acute osteomyelitis and septic arthritis, and not less than 6 months for chronic osteomyelitis) is shown in Table II. The total number of patients with adequate follow-up after completion of therapy was small. All of the chronic osteomyelitis patients had inadequate post-therapy follow-up. Although the number of the patients with acute osteomyelitis and/or septic arthritis was small, the results were impressive for the infections caused by Pseudomonas aeruginosa. This encouraging findings, however, should be confirmed by further clinical studies of this drug in comparative studies.

The adverse reactions occurred in 7 of the 11 patients. The reactions possibly or probably related to drug therapy were one each of diarrhea and pruritus, eosinophilia in 3, transient elevation of transaminases (ALT and/or AST) in 4. The type of adverse reactions seen in the additional cases were similar to those seen in earlier cases.

Conclusions: The additional information concerning the longer follow-up period of earlier cases and the additional cases provided by the applicant were evaluated by this reviewer. The conclusion reached failed to negate earlier recommendations that the favorable therapeutic results seen in this open study should be confirmed by adequate and well controlled clinical trials of this drug compared with other antibiotics approved for this indication.

Table I

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (Bone and Joint) due to Aerobic Gram-negative Organisms

Infection/Pathogen	Microbiologic Response* Number eradicated**/Number Treated Domestic	Foreign
Osteomyelitis:		
Pseudomonas aeruginosa	3/3*** (2 - acute)	0/1 (chronic)
Escherichia coli	1/1 (chronic)	-
K. pneumoniae	-	1/1 (chronic)
Enterobacter aerogenes	-	1/1 (chronic)
Morganella morganii +	-	1/1 (chronic)
E. coli + P. mirabilis	-	-
Total	4/4	3/4
Septic Arthritis (acute):		
P. aeruginosa	3/3***	-

* Microbiologic response at 10 days to 2 months after completion of therapy.
** Eradication was assumed when signs and symptoms consistent with the infection resolved.
*** One of the three patients had both acute osteomyelitis and septic arthritis.

N 50580 -2

Table II

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (Bone and Joint) due to Aerobic Gram-negataive Organisms

Infection/Organisms	Number cured*/Number treated	
	Domestic study	Foreign Study
Acute osteomyelitis:	7/8**	-
Pseudomonas aeruginosa	3/4**	-
Enterobacter cloacae	2/2	-
Escherichia coli	1/1	-
Proteus mirabilis	1/1	-
Chronic Osteomyelitis:	0/1	0/1
P. aeruginosa	-	0/1
P. aeruginosa + E. coli	0/1	-
Acute Septic Arthritis:	6/8**	0/0
P. aeruginosa	4/4**	-
E. coli	0/1	-
Enterobacter aerogenes	0/1	-
Serratia marcescens	1/1	-
P. aeruginosa + K. pneumoniae	1/1	-

**The criteria for microbioligic and clinical cures were the resolution of the signs and symptoms consistent with the infection occurred during and at post-therapy (1 month for acute osteomyelitis and septic arthritis and 6 months for chronic osteomyelitis) and/or the follow-up cultures were negative for the initial pathogen(s).

** One patient had both acute osteomyelitis and septic arthritis.

Recommendations: Aztreonam is approvable for the indication intra-abdominal infections caused by gram-negative pathogens (E. coli; Klebiella species, including K. pneumoniae, Enterobacter species including E. cloacae; Pseudomonas aeruginosa; Serratia species*, including S. marcescens; and Citrobacter species*, including C. freundii. It is also approvable for the indication gynecologic infections caused by the gram-negative pathogens E. coli; Enterobacter species*, including E. cloacae, Klebsiella pneumoniae; and Proteus mirabilis). The indication bone and joint infections is not approvable for the reasons stated above. It is recommended that the applicant be informed of the need for an adequate and well-controlled clinical study of aztreonam in the treatment of bone and joint infections. (For approval of other indications refer to MOR 11/12/85)

Note: *signifies that the organisms were the pathogen(s) in less than 10 (but more than 5) evaluable cases.

F.Min, M.D.

Orig Form 5
HFN-178
HFN-235
HFN-815 IT 3/10/86 - See also note on Group Leader's Memorandum
HFN-815/CSO
HFN-340
HFN-815/RNorton
HFN-815/MO/FM/2/28/86, js

Form 5 50-580

Group Leader's Comments on Medical Officer's Review dated November 12, 1985

Received: December 11, 1985
Review Completed: December 26, 1985

Drug Name: generic: Aztreonam
trade: Azactam

I. Summary of my concerns about this review

A. Dr. Min has chosen to use efficacy criteria for analyzing urinary tract and respiratory tract infections which have not been used by any other reviewers in the nine years I have been with FDA. They are unnecessarily harsh, they make large numbers of normally evaluable patients unevaluable, and they result in cures rates for this NDA which are not comparable with any previously approved antibiotic since they are based on quite different criteria (see detailed comments).

B. Dr. Min insists that all claims be based on controlled studies, and is unwilling to accept open studies in support of demonstrating efficacy, with one proviso. She is willing to grant claims for septicemia and skin/skin structure infections, provided that the following qualifying statement is included in the labeling: "Although data from controlled clinical trials are not available, aztreonam has been shown in uncontrolled studies to be effective in the treatment of the following indications:". This is wording that has been proposed at a Medical Officers' meeting for use in reviewing supplements, but has not been agreed upon by the Division and has not previously been suggested for use in reviewing original NDA's. None of her positions relative to controlled studies have been Division policy to date and I feel it is a very unwise precedent to unilaterally initiate them without Divisional agreement.

C. There are no summary evaluations of the results of the studies by organ system, by controlled sudies, by uncontrolled studies, or by overall results. This makes it extremely difficult to ascertain on what basis Dr. Min granted or denied claims.

II. Detailed Comments

A. Comparative Studies - pages 17-91

1. Urinary tract infections - pages 17 through 71

For reasons of her own, Dr. Min accepted as patients evaluable for efficacy only those patients who had follow-up cultures done at 4-6 weeks following the completion of therapy, provided they also had cultures done at 2-4 days during and 5-9 after therapy.

Although some reviewers have required some 4-6 week cultures to supplement the day 5 to 9 efficacy evaluation in an effort to evaluate relapse rates and the organisms causing reinfections, no reviewer since I have been with FDA has abandoned the 5-9 day post therapy culture as the basis for determining efficacy in favor of an evaluation at 4-6 weeks following therapy. Recurrence of an infection at 4-6 weeks is for more related to host factors (strictures, cysts, stones, etc.) than to efficacy of the drug (even our guidelines state this) and to me it is quite inappropriate that Dr. Min chose this as her primary criterion of efficacy. I could sympathize with a requirement for an analysis of efficacy at 5-9 days post therapy, and an analysis of relapse/reinfection at 4 to 6 weeks in cases of complicated UTI, but her present analysis is not clinically relevant. Additionally, it makes the cure rates for aztreonam non-comparable with any previously approved antibiotic.

For her evaluation, Dr. Min defined a cure as a negative culture at 2-4 days of therapy, 5-9 days following therapy, and 4-6 weeks following therapy. She divided patients who were not considered cures by her criteria into four categories; P-persistence (positive culture for some organism at 2-4 days during and/or 5-9 days post therapy - it is not clear whether a culture at 4-6 weeks p Rx was required here); RL - relapse (relapse with the original organism at 4-6 weeks); RI (E) and RI (L) - re-infection early (5-9 days) and re-infection late (4-6 weeks) [re-infection with an organism other than the original cause of the infection]; and superinfection (infection with another organism during therapy). A patient with re-infection at 5-9 days p Rx but who did not have a 4-6 weeks culture was not considered evaluable.

Although Dr. Min devotes 54 pages to the review of UTI infections, there is no overall summation of UTI results, no table of infections cured by organism, no summation of complicated versus uncomplicated infections, or IM vs IV efficacy.

2. Lower respiratory tract infections - pages 72-80

Contrary to what has been the accepted norm in the Division - a follow-up visit to evaluate efficacy 1 to 5 days after completion of therapy - Dr. Min insists on a follow-up at both 1-5 days and 3 to 4 weeks after completion of therapy for a patient to be evaluable.

If a patient had a negative culture at the 1 to 5 day follow-up but was positive at 3-4 weeks, Dr. Min called these patients a failure in contrast to the sponsor's finding of a "microbiological cure with relapse." Dr. Min's stringent criteria resulted in 2/3 of the patients being unevaluable.

Again, there is no overall summary and evaluation of bacteriological and clinical results by organism.

3. Ob-Gyn infections - pages 86-87.

There is no overall evaluation of the bacteriological and clinical results by organism.

4. Comparative Studies

On the concluding pages 90 and 91, there is no overall summary and evaluation of the bacteriological and clinical results by organism.

B. Non-comparative studies - pages 91 to 125

1. Urinary tract infections

pages 91-95. This was an open study of urinary tract infections resistant to other antibiotics. Again Dr. Min insists on a 4-6 weeks culture for a patient to be evaluable. Since that study is non-comparative she feels it has limited meaning. I disagree with both conclusions.

2. Osteomyelitis

Dr. Min continues to insist that an adequate and well-controlled clinical study be done for each indication. Is this a proper requirement?

3. Intra abdominal infections, pages 101-105.

There are no conclusions and recommendations

4. Ob-Gyn infections, pages 105-107

There are no conclusions and recommendations

5. Septicemia, pages 107-111

There are no conclusions and recommendations

6. Lower respiratory tract

The uncommonly low cure rates for aztreonam in the treatment of lower respiratory tract infections, especially those due to Pseudomonas, is due to Dr. Min's inclusion of patients with cystic fibrosis based on clinical improvement (it is generally acknowledged that they rarely have bacteriological cure). It gives a very false impression of cure rates to list cystic fibrosis Pseudomonas persistence as failure. Cystics should be analyzed separately from other patients.

Pages 120-121. I assume that the low cure rates for Pseudomonas are due to the inclusion of cystic fibrosis patients.

7. Urinary Tract Infections

p. 122. Again, evaluable patients are only those that include a 4-6 weeks post therapy follow-up.

C. Safety

p. 126 is apparently the start of the safety analysis, but is untitled. Is this the safety for the open studies (which it follows) or the entire NDA?

p. 127 Deaths. Are these for open studies, or the entire NDA?

D. Overall Conclusions and Recommendations

p. 128 There is no overall summary of efficacy results, making it extremely difficult to assess the basis on which claims were granted.

p. 129 Dr. Min feels that controlled studies must be done for all indications. She is willing to grant claims for septicemia and skin/skin structure infections based on uncontrolled studies, provided that the following qualifying statement is included in the labeling: "Although data from controlled clinical trials are not available, aztreonam has been shown in uncontrolled studies to be effective in the treatment of the following indications:". This wording has not been used previously in approved labeling and has only tentatively been proposed for the review of supplements - not labeling. Its use without Divisional agreement is bad precedent.

Group Leader's Conclusions and Recommendations

I can not concur in Dr. Min's review because of the reasons outlined in my own review. Accepting the review as it is will make its results and conclusions non-comparable with any other NDA the Division has approved. On the other hand, because of the long time that aztreonam has been under review in the Division, Squibb is more than likely willing to have an approval based on such stringent and atypical criteria solely for the ability to at least finally market the product. These unrepresentatively low cure rates will undoubtedly pose a problem in advertising.

I will defer to Dr. Tabor's conclusions.

George R. Stanley, M.D.
Group Leader, DAIDP

MEDICAL OFFICER'S REVIEW OF FORM 5 (50-580)

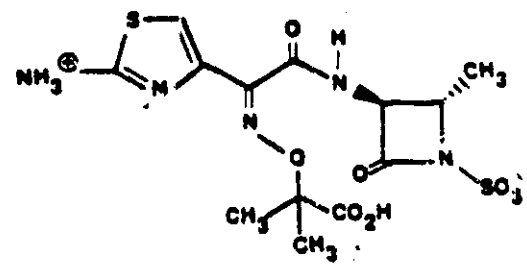
November 12, 1985

Applicant: E.R. Squibb and Sons, Inc.

Name of Drug:

Trade - Azactam for Injection
Code - SQ26,766
Generic - Aztreonam
Chemical - (Z)-2-[[[(2-amino-4-thiazolyl)[[(2S,3S)-2-methyl-4-oxo-1-sulfo-3-azetidinyl[carbamoyl]methyleneamino]oxy]-2-ethylpropionic acid

Structure:



Category of Drug: Synthetic antibiotic (monocyclic beta-lactam, monobactam)

Proposed Indications by the Applicant: For the treatment of the following infections caused by susceptible gram-negative microorganisms, including Escherichia coli, Enterobacter species, Klebsiella spp. including K. pneumoniae and K. oxytoca, Proteus mirabilis, P. vulgaris, Morganella morganii (formerly P. morganii), Providencia species including P. stuarti and P. rettgeri), Pseudomonas species including Ps. aeruginosa, Serratia marcescens, Neisseria gonorrhoeae including beta-lactamase producing or non-producing strains, Haemophilus influenzae including ampicillin- resistant and other penicillinase-producing strains, Citrobacter species, and some strains of Acinetobacter calcoaceticus

Urinary tract infections, including pyelonephritis and cystitis (initial and recurrent) and asymptomatic bacteriuria
Lower respiratory tract infections, including pneumonia and bronchitis.
Bacteremia/septicemia
Bone and joint infection
Skin and skin-structure infections, including those associated with postoperative wounds, ulcers, and burns.
Intra-abdominal infections, including peritonitis.
Gynecologic infections, including pelvic inflammatory disease, endometritis, and pelvic cellulitis.
Acute gonorrheal infection (uncomplicated urogenital or anorectal)

Azactam has proven highly effective in therapy for most patients with serious urinary tract infections caused by multi-resistant gram-negative aerobic pathogens. AZACTAM is indicated for adjunctive therapy to surgery in the management of infections caused by susceptible organisms, including abscesses, infections complicating hollow viscus perforations, cutaneous infections of serous surfaces. AZACTAM is effective against most of the commonly encountered gram-negative aerobic pathogens seen in general surgery.

Concurrent Therapy: Concurrent initial therapy with other antimicrobial agents and AZACTAM is recommended before the causative organism(s) is known in seriously ill patients who are also at risk of having an infection due to gram-positive aerobic pathogens. If anaerobic organisms are also suspected as etiologic agents, therapy should be initiated using an antianaerobic agent concurrently with AZACTAM.
Following identification and susceptibility testing of the causative organism(s), appropriate antibiotic therapy should be continued.

Patients may be benefit from concurrent use of aztreonam and an aminoglycoside. These agents are synergistic in vitro against most strains of Pseudomonas aeruginosa, many strains of Enterobacteriaceae, and other gram-negative aerobic bacilli. However, this enhanced activity is not predictable. If such concurrent therapy is considered in patients with serious infections, susceptibility test should be performed to determine the activitity of the drugs in combination

Dosage Form and Route of Administration: 1 gram aztreonam with approximately 780 mg L-arginine/vial, for intravenous and intramuscular use after constitution.

Related IND and NDA: IND 18,554 (Aztreonam; E.R. Squibb and Sons, Inc.); no related NDA.

Date of NDA Submission: June 1, 1983 (Preclinical data-microbiological and animal toxicological- and pharmacokinetic data), October 10, 1983 and November 30, 1983 (Manufacturing and control data); December 28, 1983 (Clinical data).

Microbiologist's Reviews: Refer to Ms. Eckert's reviews dated 12/9/83 and 8/9/84. She stated that the Microbiology section of the package insert in the proposed labeling is satisfactory from the microbiologis's viewpoint.

Pharmacologist's Reviews: Refer to Dr. Alam's reviews dated June 21, 1984 and December 18, 1984.

Pharmacokinetic studies of aztreonam in animals:

Peak serum concentrations were achieved about 10-20 minutes after parenteral dosing and 40-50% of the dose was excreted in the urine in mice, monkeys and rats. In rodents about 3% of the dose was recovered in the bile during a 2-hour period. A radiolabelled study of aztreonam in rats showed that the drug is well-distributed in tissues. The concentrations of the drug in tissues (kidney, liver, urinary bladder and meninges) were higher than in serum after single intramuscular injection of 50 mg/kg doses. Significant concentrations of the drug were also detected in cerebrospinal fluid (CSF), placentas, fetuses and amniotic fluid. It is excreted in milk of lactating rats. The serum half-lives of parenterally administered drug (IV, IM, or SC) were about 1 hour in dogs and between 1 and 6 hours in monkeys. In monkeys, SQ 26,992 was the only major metabolite identified in the urine. None of four metabolites detected possesses antimicrobial activity.

Toxicology study in animals:

In the rat SC toxicity study the "no-effect" dose of the arginine blend aztreonam was found to be 150 mg/kg. Increased liver weights without histopathological changes were seen in the high- (2400 mg/kg) and mid-(600 mg/kg) dosed animals. The increased kidney weights in the high-dose group, however, was accompanied by mild vacuolation in the renal tubular epithelial cells. Similar hepatic and renal changes were observed in the 30-day IV dog study indicating that the kidney and possibly the liver appear to be the target organs for toxicity of this drug. In the Japanese study in rats, moderate centrolobular hypertrophy of hepatocytes and slight to moderate vacuolation of the tubular epithelium of the kidneys were seen in the high-(2000 mg/kg) and mid-(750 mg/kg) dose animals. The "No effect" dose in this study was 100 or possibly 270 mg/kg.

Pharmacokinetic Studies in Humans: Several pharmacokinetic studies of single or multiple doses of aztreonam administered intravenously were performed in healthy volunteers as well as in patient populations. The results of the studies are under review by the Pharmacokinetic Branch. A preliminary review of the data indicated that the serum and tissue levels of aztreonam attained are significantly higher than the MICs of aerobic Gram-negative pathogens, including Pseudomonas aeruginosa. The pertinent pharmacokinetic parameters of aztreonam in healthy volunteers and in those with renal dysfunction were reported by Swabb et al. (Amer J Med 1985;78(suppl 2A):11-18) The following Tables and Figures were copied from the report:

TABLE I Serum and Urinary Concentrations (μg/ml, mean ± SEM) of Aztreonam after 0.5 g Parenteral or Oral Dose in Healthy Volunteers*

Time after Dosing	Route of Administration		
	Intravenous	Intramuscular	Oral Solution
Serum			
5 minutes	56 ± 3	4.8 ± 1.0	—
1 hour	23 ± 1	22 ± 2	0.11 ± 0.02
4 hours	6.7 ± 0.2	8.9 ± 0.5	0.11 ± 0.02
6 hours	2.9 ± 0.2	3.8 ± 0.3	0.08 ± 0.02
8 hours	1.3 ± 0.1	1.7 ± 0.2	<0.04
12 hours	0.28 ± 0.04	0.30 ± 0.05	<0.04
Area under the curve (μg × hour/ml)	94 ± 2	84 ± 3	0.45 ± 0.08
Urine			
0–2 hours	1,400 ± 200	520 ± 190	2.9 ± 0.7
4–6 hours	330 ± 57	420 ± 87	6.6 ± 1.4
8–12 hours	80 ± 8	27 ± 8	1.4 ± 0.2
16–24 hours	1.9 ± 0.4	1.3 ± 0.3	0.31 ± 0.06
Percent recovery	69 ± 2	62 ± 4	0.7 ± 0.1

*Numbers of different subjects receiving aztreonam intravenously, intramuscularly, and orally were six, six, and 15, respectively. (Data from [3–5] with permission.)

TABLE II Serum and Urinary Concentration (μg/ml, mean ± SEM) of Aztreonam after 1 g and 2 g Parenteral Dose in Healthy Volunteers*

Time after Dosing	1 g Dose		2 g Dose
	Intravenous	Intramuscular	Intravenous
Serum			
5 minutes	125 ± 4	8.6 ± 2.3	242 ± 20
1 hour	49 ± 2	46 ± 3	91 ± 6
4 hours	13.2 ± 0.3	16.4 ± 0.6	26 ± 2
6 hours	6.0 ± 0.3	9.2 ± 0.4	13 ± 1
8 hours	2.7 ± 0.1	3.5 ± 0.2	6.0 ± 0.6
12 hours	0.51 ± 0.02	0.86 ± 0.09	1.2 ± 0.1
Area under the curve (μg × hour/ml)	191 ± 5	180 ± 4	379 ± 23
Urine			
0–2 hours	3,000 ± 1,200	1,200 ± 320	6,800 ± 1,100
4–6 hours	720 ± 190	640 ± 200	1,800 ± 520
8–12 hours	70 ± 10	140 ± 28	180 ± 56
16–24 hours	2.8 ± 0.5	5.0 ± 2.7	9.8 ± 3.1
Percent recovery	74 ± 3	69 ± 3	66 ± 3

*Six different subjects received a 1 g intravenous dose, six subjects a 1 g intramuscular dose, and six subjects a 2 g intravenous dose. (Data from [3,5] with permission.)

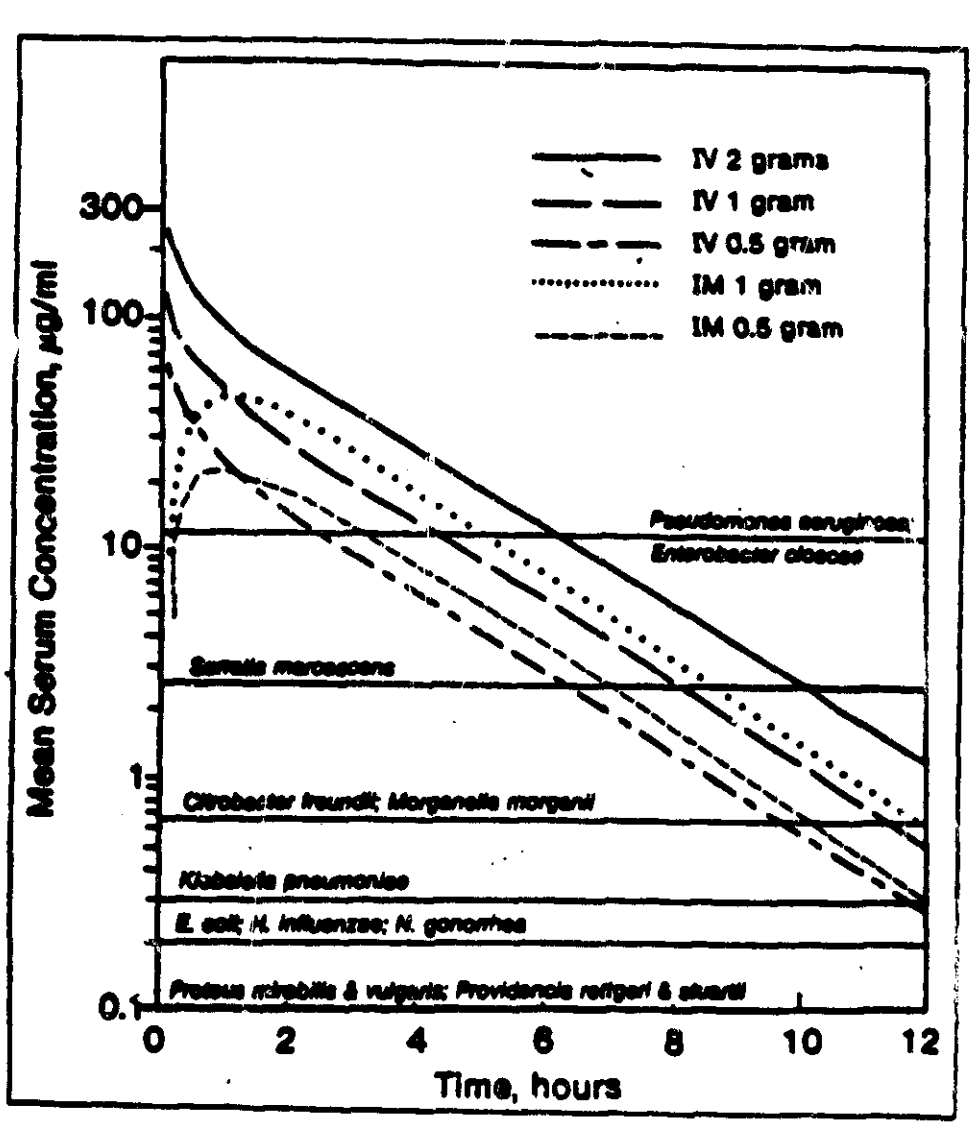
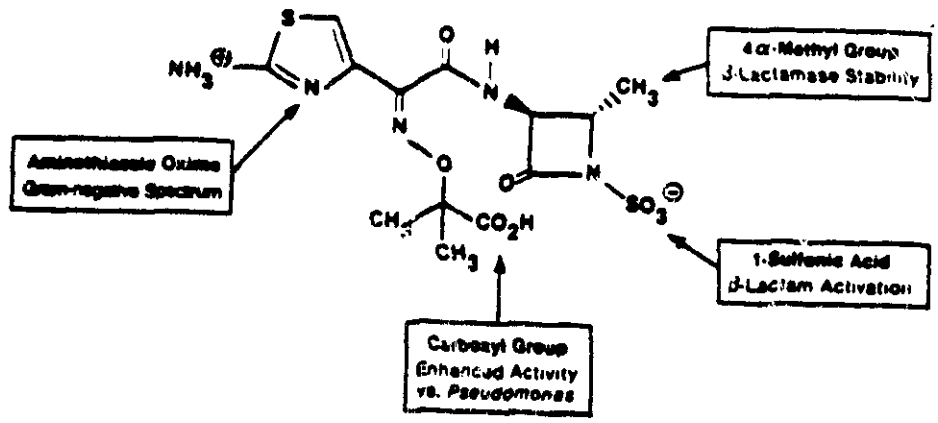


Figure 1. Comparison of pharmacokinetic profile of aztreonam in serum of healthy subjects with antibacterial activity of aztreonam in vitro. (Adapted from [2,3] and [33] with permission.)

From Swabb, RA, Amer J Med 1985; 78 (S 2A):11-18

Microbiology:

Aztreonam is the first of the synthetic monocyclic beta-lactams (monobactams). It is extremely active against aerobic gram-negative bacteria even in low concentrations, but relatively inactive against gram-positive and anaerobic micro-organisms, as shown in Table I. It interacts with certain penicillin-binding proteins of these organisms and thus interferes with the biosynthesis of bacterial cell walls. It is highly resistant to enzymatic hydrolysis by beta-lactamases, similar to the aminothiazolyl cephalosporins and moxalactam, and demonstrates a high degree of stability against plasmid-mediated gram-negative lactamases as well. It does not induce production of chromosomally-mediated enzymes.
The structure-activity relationship in the aztreonam molecule is depicted in Figure 1.



The potent antibacterial activity of this drug against aerobic gram-negative clinical isolates as compared to other antibiotics are shown in Tables II and III. The in vitro data indicate that antimicrobial activity of aztreonam is equivalent to the third generation cephalosporins against gram-negative bacteria, but it has little or no activity against gram-positive bacteria and anaerobic bacteria. The overall activity of aztreonam against E. coli and Klebsiella is similar to that of cefotaxime and moxalactam. Its' activity against all Proteus species is superior to that of these antibiotics. The antipseudomonas activity of aztreonam is also superior to moxalactam. Only Enterobacter and a rare Citrobacter are resistant. Its activity against multiply resistant Enterobacteriacese was reported to be similar to that of the third generation cephalosporins (Acar et al.). As shown in Table V, synergistic activity between aztreonam and aminoglycosides was demonstrated against the majority of P. aeruginosa strains, but not against the majority of Acinetobacter species (Sykes et al., 1985). Stutman and his associates also reported a similar finding that the tobramycin combination was synergistic (62%) against P. aeruginosa. Their experiments also showed that combinations of aztreonam with cefoxitin or clindamycin are synergistic, particularly against E. coli, Klebsiella-Enterobacter spp. and Shigella spp. Wu et al. studied the effect of aztreonam in combination with antipseudomonal penicillins (azlocillin and piperacillin) on P aeruginosa. The results of their study indicated that aztreonam has little synergistic effect against this bacteria when used in combination with the antipseudomonal penicillins. Antagonism, however, was not seen. They reported that aztreonam appears to be more stable than the penicillins in the presence of the chromosomally mediated class 1d beta-lactamase produced by this microorganism. Among the non-Enterobacteriaceae, aztreonam is highly active against the Neisseria and Haemophilus species. As shown in Table IV, the ranges of the MIC and MBC of Enterobacteriaceae tested are similar.

Table I

Antibacterial Activity of Aztreonam

Organisms (No. of Strains)	MIC Range (ug/ml)	MIC needed to inhibit 90% of Strains
Enteric Gram-negative Bacilli:		
Bacteroides fragilis (8)	100 - 100	100
Citrobacter freundii (25)	0.1 - 50	0.7
Enterobacter aerogenes (13)	0.1 - 50	33.3
E. cloacae (29)	0.1 - 50	12.5
Escherichia coli (79)	0.1 - 0.8	0.2
Klebsiella pneumoniae (68)	0.1 - 100	0.3
Morganella morganii (19) (formerly P. morganii)	0.1 - 1.6	0.6
Proteus mirabilis (25)	0.1 - 0.1	0.1
P. vulgaris (11)	0.1 - 0.1	0.1
Providencia rettgeri (6) (formerly P. rettgeri)	0.1	0.1
Providencia stuartii (15)	0.1 - 0.1	0.1
Salmonella sp. (25)	0.1 - 0.8	0.3
Serratia marcescens (13)	0.1 - 6.3	1.6
Shigella sp. (25)	0.1 - 12.5	5.7
Other Gram-negative Bacilli:		
Acinetobacter calcoaceticus (25	1.6 - 100	58.3
Haemophilus influenzae (18) (ampicillin-susceptible)	0.1 - 0.2	0.2
H. influenzae (18) (ampicillin-resistant)	0.1 - 0.2	0.2
Pseudomonas aeruginosa (61)	0.2 - 50	12.0
Gram-positive Cocci:		
Staphylococcus aureus (12)	100	100
Streptococcus pyogenes (11)	12.5 - 100	12.5
S. pneumoniae (11)	50 - 100	100
S. fecalis (12)	100	100
Gram-negative Cocci:		
Neisseria gonorrhoeae (20)	0.1 - 0.4	0.2

From: Sykes RB et al. Antimicrob Ag Chemother 1982; 21:85-92
Inoculum size - 5×10^5 CFU

Table II

Antimicrobial Activity Against Clinical Isolates*

Organism (No. of strains)	MIC_{90}(ug/ml) Aztreonam	Cefotaxime	Moxalactam	Gentamicin
E. coli (50)	0.2	0.2	0.4	5.0
K. pneumoniae (51)	0.2	0.1	0.6	1.6
P. mirabilis (25)	0.1	0.1	0.2	5.0
Proteus (indole-positive), Providencia sp. (39)	0.1	2.2	0.4	17.0
Salmonella sp. (24)	0.3	0.3	0.3	2.4
Citrobacter sp. (24)	0.7	0.6	0.4	1.5
Enterobacter sp. (43)	15.0	25.0	15.0	2.3
Serratia sp. (49)	0.9	0.8	2.7	47.0
Pseudomonas sp. (50)	15.0	47.0	46.0	3.1
Acinetobacter sp. (20)	66.0	23.0	83.0	1.6
Neisseria gonorrhoeae (19)	0.2	0.1	0.1	3.4
Haemophilus influenzae ampicillin-sensitive (10)	0.2	0.1	0.1	3.1
H. influenzae ampicillin-resistant (18)	0.2	0.1	0.1	3.0

-From Sykes RB et al. Amer J Med 1985, 78(2A):2-10
*Inoculum size - 5×10^5 CFU

Table III

Comparative Activity of SQ and B-lactam Compounds

Organism (No. of strains)	MIC (ug/ml) Range/MIC_{90} (ug/ml) Aztreonam	Cefotaxime	Moxalactam
Klebsiella oxytoca (14)	0.05 - 0.8 (0.8)	0.05 - 0.4	-
Neisseria menigitidis (5)	0.012-0.025 (0.025)	0.025-0.05 (0.05)	0.025-0.05 (0.05)
Aeromonas hydrophila (10)	0.01 - 0.5 (0.1)	0.01 - 0.4 (0.1)	- -
Pasteurella multocida	0.02 - 0.1 (0.1)	0.02 - 0.1 (0.1)	-
Yersinia enterocolitica (5)	0.02 - 3.1 (3.1)	0.02 - 0.8 (0.8)	-

From Neu HC et al. J Antimicrob Chemother 1981; 8 (Suppl E): 111-22
Inoculum size - 10^5 CFU

Table IV

Antimicrobial Activity of Aztreonam Against Gram-negative Isolates

Orgnaisms	Mean MIC (ug/ml)	MIC Range	MBC Range*
E. coli (12)	0.07	0.03 - 0.5	0.03 - 0.5 (1)
Klebsiella spp. (12)	0.04	0.015 - 0.12	0.03 - 0.12 (0)
Enterobacter spp. (12)	0.05	0.015 - 1.0	0.015 - 1.0 (0)
S. marcescens (10)	0.19	0.06 - 1.0	0.06 - 2.0 (0)
P. mirabilis (10)	0.009	0.008 - 0.03	0.008 - 0.12 (2)
Shigella spp. (12)	0.04	0.008 - 0.12	0.008 - 0.25 (0)
Salmonella spp. (12)	0.07	0.03 - 0.25	0.03 - 0.5 (0)
P. aeruginosa (18)	1.7	0.25 - 32	0.25 - 32 (2)

From Stutman HR et al. Antimicrob Ag Chemother 1984, 25:212-5
Values in parentheses indicate the number of strains for which the MBCs are 2X the MICs.

Table V

Interaction of Aztreonam with Aminoglycosides Against Gram-negative Bacteria

Organism	Strains Synergistically Inhibited*/No. Tested		
	Gentamicin	Tobramycin	Amikacin
P. aeruginosa	19/27 (70.4%)	24/26 (88.9%)	22/27 (81.5%)
Acinetobacter sp.	6/25 (24.0%)	7/25 (28.0%)	8/25 (32.0%)

From Sykes RB et al. Amer J Med 1985, 78 (Suppl 2A): 2-10
*Synergy defined as fractional inhibitory concentration of 0.5 or less.

Controlled Clinical Studies

Protocol 18554-10: Comparison of Aztreonam(IM) and Spectinomycin (IM) in the Treatment of Acute Uncomplicated Gonorrhea.

Four principal investigators, three in the U.S. and one U.S. navy physician in the Philippines, participated in the open randomized comparative study of aztreonam vs. spectinomycin in the treatment of acute uncomplicated gonococcal infections of the genito-urinary tract. Patients with a presumptive diagnosis of gonococcal urethritis/cervicitis or anorectal gonococcal infection were entered into the study. The presumptive diagnosis was based on the demonstration of typical gram-negative intracellular diplococci in gram stained exudate from appropriate sources using adequate procedures. Confirmation of the diagnosis was made by the isolation of Neisseria gonorrhoeae. Susceptibility of the isolates was determined by the MICs. In female patients, cultures for Monilia, Trichomonas, and Gardenella were obtained. However, cultures for Chlamydia and Ureaplasma were not done.

Patients were randomly assigned to receive either a single dose of aztreonam (1 g) or spectinomycin (2 g) intramuscularly, according to randomization codes. Routine hematology and blood chemistries were done only for the patients in the aztreonam group. Follow-up clinical and bacteriological examinations and repeat clinical laboratory tests were done between 3 and 7 days after completion of the single dose therapy. The applicant provided the summaries of individual investigators' studies, and the results of these studies were pooled in its final analyses. Since the patient populations and the methods of these studies varied, as shown in the following tables, this reviewer did not pool the data in her analyses. Two investigators (Drs. Harrison and Lutz) treated a substantial number of patients. One investigator (Dr. Slutkin) did not record the MICs of clinical isolates. Bacteriological cure was defined as eradication of N. gonorrhoeae at the designated follow-up period. Clinical cure was defined as subsidence of clnical symptoms and signs attributed to gonococcal infections.

The total numbers of patients entered into the studies by each investigator, and the number of evaluable patients for efficacy analyses varied as, shown in the succeding Tables. The overall bacteriological cure rate ranged from 82 % to 100 % in the males, and was 100 % in the female patients. These cure rates were comparable to those in the control groups treated with spectinomycin. The numbers of patients with penicillinase producing N. gonorrhoeae (PPNG) infections treated either with aztreonam (23) or spectinomycin (24) were small. The cure rates, nevertheless, were similar to those in non-PPNG infections, and were comparable in the two treatment groups. In none of the failure cases was an increase of the MICs noted. One investigator (Slutkin) did not record the MICs of the clinical isolates in all of his patients. Bacteriological cures were seen in all of 8 males in the aztreonam group and all of 11 males in the spectinomycin group. In other studies, the MICs of the clinical isolates were not recorded in a few patients. The bacteriological cure rates in those patients were similar to those seen in the Slutkin study. Instances of post-gonococcal urethritis were also comparable in the test and control groups.

Adverse reactions noted were primarily local reactions at the sites of intramuscular injections. Pain at the injection site was common in both treatment groups: 40.7% of 258 patients in the aztreonam group and 46.5% of 254 patients in the spectinomycin group. In the majority of the patients, the pain was slight. Induration and/or erythema were seen in 1.9% and 5.9% of the aztreonam group and the spectinomycin group, respectively. Other reactions obseved were: one case each of nausea and vomiting, and fatigue and lethargy in the aztreonam group, and one case of urticaria in the spectinomycin group. No laboratory abnormalities attributable to the test or the control drug were noted.

Conclusions: The results of the multicenter, randomized comparative study of single, one-gram doses of aztreonam (IM) vs. single two-gram doses of spectinomycin (IM) indicate that aztreonam is as effective as spectinomycin, an effective antibiotic for PPNG, in the treatment of acute, uncomplicated gonococcal urethritis in males and cervicitis/urethritis/proctitis in females. The numbers of patients treated for anorectal N. gonorrhoeae infections, however, are small: 10 females and 3 males in the aztreonam group and 7 females and 4 males in the spectinomycin group. Bacteriological cures were seen in all of 10 females in the test drug group, and in 5 of 7 in the control drug group.

Protocol 18554-10: Comparison of Aztreonam(IM) and Spectinomycin (IM) in the Treatment of Acute Uncomplicated Gonorrhea

Investigator's Name and ID Number: Wm. O. Harrison (Naval Regional Medical Center, San Diego, CA); 6221

	Aztreonam	Spectinomycin
Total Number of Patients Entered into the Study	56	54
Number of Patients Excluded from the Efficacy Evaluation	14	16
Reasons for Exclusions:		
No pathogens isolated	0	7
Repeated sexual contact after therapy	1	0
No post-therapy follow-up	5	2
Other antimicrobial therapy	0	1
No susceptibility testing	8	6
Number of Evaluable Patients	42	38
Demographic Characteristics of Evaluable Patients:		
Sex		
Male	42	38
Age (years)		
Range	18 - 39	17 - 34
Mean	23.5	24.5
Age not recorded	12	11
Race		
Black	11	6
Caucasian	31	32
Infection Site		
Urethra	42	38
Organisms		
penicillinase producing (PP) strains	23	24
non-penicillinase producing (NPP) strains	19	13
not tested	0	1
MICs (ug/ml)		
PP strains	0.0005 - 0.125	1 - 32
Non-PP strains	0.004 - 2	1 - 32

Bacteriological and Clinical Response

	Aztreonam	Spectinomycin
Bacteriological and Clinical cure	42/42 (100.0%)	38/38 (100.0%)

Protocol 18554-10: Comparison of Aztreonam(IM) and Spectinomycin (IM) in the Treatment of Acute Uncomplicated Gonorrhea

Investigator's Name and ID Number: H. Handsfield, M.D. (Harborview Medical Centerter, Seattle, WA; 6359

	Aztreonam	Spectinomycin
Total Number of Patients Entered into the Study	40	39
Number of Patients Excluded from the Efficacy Evaluation	8	14
Reasons for Exclusions:		
No pathogens isolated	2	3
No post-therapy follow-up	2	6
Culture site not stated	4	5
Number of Evaluable Patients	32	25
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	4	6
Male	28	19
Age (years)		
Range		
Females	16 - 26	16 - 38
Males	17 - 46	15 - 48
Mean		
Females	20.5	21.2
Males	26.9	28.8
Race		
Black	8	6
Caucasian	23	15
Other	1	3
Not recorded	0	1
Infection Site		
Females:		
Cervix only (C)	1	2
Rectum only (R)	1	0
C + R	1	2
C + R + pharynx	1	1
Urethra only	0	1
Males:		
Urethra only	25	17
Urethra + rectum	2	1
Urethra + pharynx	1	1

Investigator No. : 6359

Bacteriological and Clinical Response

	Number cured/Number treated	
	Aztreonam	Spectinomycin
Females:		
Cervix only (C)	1/1	2/2
Rectum only (R)	1/1	0/0
C + R	1/1	2/2
C + R + pharynx	1/1	0/1
Urethra only	0/0	1/1
Total	4/4	5/6
Males:		
Urethra	22/25 (88.0%)	15/17 (88.2%)
Urethra + rectum	1/2	1/1
Urethra + pharynx*	0/1	0/1
Total	23/28 (82.1%)	16/19 (84.2%)

* Failure involves pharyngeal infection.

Protocol 18354-10: Comparison of Aztreonam(IM) and Spectinomycin (IM) in the Treatment of Acute Uncomplicated Gonorrhea

Investigator's Name and ID Number: B. Lutz, M.D. (New Orleans, LA), 6360

	Aztreonam	Spectinomycin
Total Number of Patients Entered into the Study	158	155
Number of Patients Excluded from the Efficacy Evaluation	46	44
Reasons for Exclusions:		
No pathogens isolated	41	36
No post-therapy follow-up	5	8
Number of Evaluable Patients	112	111
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	75	77
Male	37	34
Age (years)		
Female:		
Range	17 - 37	18 - 32
Mean	23.6	22.6
Male:		
Range	18 - 39	17 - 54
Mean	24.6	24.4
Race		
Black	104	104
Caucasian	8	4
Not recorded	0	3
Infection Site		
Females:		
Cervix only (C)	17	22
C + urethra	51	50
C + rectum	0	1
C + urethra + rectum	7	4
Males:		
Urethra only	37	34

Bacteriological Response

	Number cured/Number treated	
	Aztreonam	Spectinomycin
Females:		
Cervix only (C)	17/17 (100.0%)	21/22 (95.5%)
Cervix + urethra	51/51 (100.0%)	49/50 (98.0%)
Cervix + urethra + rectum	7/7	3/4
cervix + rectum	0/0	1/1
Total	75/75 (100.0%)	74/77 (96.1%)
Males:		
Urethra	35/37 (94.6%)	34/34 (100.0%)

Clinical Response

	Number cured/Number treated	
	Aztreonam	Spectinomycin
Females:		
Cervix only	11/13 (84.6%)	13/15 (86.7%)
Cervix + urethra	27/31 (87.1%)	29/33 (87.9%)
Cervix + urethra + rectum	4/4	2/3
Cervix + rectum	1/1	1/1
Total	43/49 (87.8%)	45/52 (86.5%)

Note: Fifty-one patients (26 in the aztreonam group and 25 in the spectinomycin group) were not evaluable for the clinical response since the clinical symptoms and signs were not recorded prior to the initiation of antimicrobial therapy.

Males:		
Urethra	36/37 (97.3%)	32/34 (94.1%)

Protocol 18554-10: Comparison of Aztreonam and Spectinomycin in the Treatment of Acute Uncomplicated Gonorrhea.

Investigator's Name and ID Number: G. Slutkin, M.D. (SF General Hospital, San Francisco, CA); 6361*

	Aztreonam	Spectinomycin
Total Number of Patients Entered into the Study	14	16
Number of Patients Excluded from the Efficacy Evaluation	6	3
Reasons for Exclusions:		
No pathogens isolated	4	1
No post-therapy follow-up	0	1
Culture sites not stated	2	1
Number of Evaluable Patients	8	13
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	0	2
Male	8	11
Age (years)		
Range	30 - 41 (M)	23 - 50 (F) 25 - 41 (M)
Mean	33.9 (M)	36.5 (F) 28.5 (M)
Race		
Caucasian	8	12
Not recorded	0	1
Infection Site		
Females:		
Cervix only (C)	0	2
Males:		
Urethra only	7	8
Rectum only	1	3

* This investigator did not record the MICs of the clinical isolates.

Investigator Number : 6361

Bacteriological Response*

	Number cured/Number treated	
	Aztreonam	Spectinomycin
Females:		
Cervix only (C)	0/0	2/2
Males:		
Urethra only	7/7	8/8
Rectum only	1/1	3/3
Total	8/8 (100.0%)	11/11 (100.0%)

*The MICs of the test and the control drugs were not recorded.

Clinical Response

	Number cured/Number treated	
	Aztreonam	Spectinomycin
Females:		
Cervix only	0/0	0/0
Males:		
Urethra only	7/7	8/8
Rectum only	0/0	3/3
Total	7/7	11/11 (100.0%)

Protocol 18554-15: Comparative Study of Aztreonam (IM) vs. Amoxicillin (P.O.) in The Treatment of Acute, Uncomplicated Urinary Tract Infection (Cystitis).

This is a multicenter, open-label, randomized study of aztreonam in the treatment of 'cystitis', in which the efficacy and safety of a single I.M. dose of aztreonam was compared to a 10-day course of oral amoxicillin.

In this study, the diagnosis of acute uncomplicated cystitis was made by the presence of clinical findings compatible with lower urinary tract infection (UTI) and significant bacteriuria ($\geq 10^5$ CFU/ml) in the absence of urinary tract abnormalities. Patients with a history of 2 or more episodes of UTI were excluded. No invasive or noninvasive localization studies were performed. Susceptibility testing of clinical isolates was performed using the disc method. Serotyping of E. coli was not done.

The dosages of aztreonam and amoxicillin used were 1 gm and 250 mg q 8 h , respectively.

Ten principal clinical investigators at 10 clinical centers in the US entered a total of 153 patients into the study. Six of the ten investigators entered at least one evaluable patient for each treatment group. Fifty six patients were excluded from the efficacy evaluation for a variety of reasons, as indicated in Table I. The demography of the patient population in the two treatment groups was comparable in respect to age, sex, race, and the duration of urinary symptoms prior to therapy. Escherichia coli (90.6%) was the predominant pathogen, as expected from such a study population. The number of patients evaluated by this reviewer differs slightly from that by the applicant, since this reviewer excluded a few patients who did not meet the criteria for a clinical diagnosis of uncomplicated UTI and those with the UTI caused by resistant pathogens, and evaluated those patients who had follow-up urine cultures on both day 4 and at 4-6 weeks after completion of therapy.

The bacteriological response seen in the test and control drug groups is presented in the following Table II. The results of the multicenter domestic study suggest that a single intramuscular dose of aztreonam is less effective than the conventional ten-day course of oral amoxicillin; the overall bacteriological cure rates for susceptible gram-negative pathogens were 75.5% (40/53) in the aztreonam group and 89.5% (34/38) in the amoxicillin group, at 4-6 weeks after completion of therapy. The failures were due to persistence (18%) or relapse (5%) in the aztreonam-treated patients, and were due to relapse (7.5%) in the amoxicillin-treated patients. The clinical response (resolution or improvement of clinical signs and/or symptoms) was seen in 75% and 88%, respectively, in the test and the control group. The bacteriological cure rates for UTI due to E. coli (a major pathogen) were 76% (38/50) in the aztreonam group and 89% (31/35) in the amoxicillin group. The clinical cure rates were 74% and 87%, respectively. These differences, however, do not appear to reach statistical significance. Since serotyping of E. coli was not done, it cannot be ascertained whether recurrence was due to relapse or reinfection caused by a different serotype. The relapse and reinfection rates in the two treatment groups were similar. The urinary pathogens causing relapse and re-infection were susceptible to aztreonam, but were resistant to amoxicillin in the majority of the cases. The number of the patients with UTI caused by non-E. coli was quite small.

To assess the safety of aztreonam, routine hematology, blood chemistry and urinalysis were performed prior to and after completion of therapy. Of the 153 patients who were entered into the study, 78 patients received aztreonam and 75 patients received amoxicillin. Four patients (5%) in the aztreonam group and 9 patients (12%) in the amoxicillin group had adverse reactions. One patient in each treatment group had more than one adverse reaction. In six patients treated with amoxicillin, the drug was discontinued, but in none treated with aztreonam. No clinically significant laboratory aberrations were seen in both treatment groups. The adverse reactions reported were as follows:

Adverse Reaction	Aztreonam	Amoxicillin
Rash	0	2
Urticaria	0	1
Diarrhea	0	2
Nausea	1	1
Vomiting	1	0
Halitosis	1	0
Breast tenderness	1	0
Dizziness	1	0
Headache	1	0
Chest pain	1	0
Vaginitis	0	1

Conclusions: The results of the domestic, multicenter, randomized, controlled study of single doses of aztreonam (IM) vs. conventional 10 day courses of amoxicillin (oral) in the treatment of acute uncomplicated lower urinary tract infections (cystitis) indicate that single doses of aztreonam appear to be less effective than the conventional amoxicillin therapy. The safety of the two treatments is comparable. Single-dose antimicrobial therapy is currently recommended by a few physicians for the treatment of urinary tract infections, specifically for acute, uncomplicated lower urinary tract infections (cystitis). However, the cure rates seen in this study appear to be less than optimum. There have been no other clinical studies in which the safety and efficacy of single doses of aztreonam were compared to those of other antimicrobials for the treatment of the acute, uncomplicated lower urinary tract infection. Such studies are needed to ascertain the role of this new monobactam in the management of uncomplicated lower urinary tract infection, for which many effective antimicrobials are available. At this time, the approval of single-dose aztreonam therapy for the proposed indication is considered premature. The higher bacteriological and clinical cure rates were reported by the applicant: 84% (47/56) and 93% (52/56), respectively, in the aztreonam group, and 93% (42/45) and 96% (43/45), respectively, in the amoxicillin group. The higher cure rates might partly be attributable to the applicant's evaluation of the responses at a shorter follow-up period (5-9 days) after completion of therapy and to the inclusion of late relapse as cure.

Table I

Protocol 18554-15

Total Number of Principal Investigators : 10

Investigators' ID Numbers: 0464*: 2891*; 3053; 4147; 4318; 5023*; 6236; 6360; 6283; 7544*

	Aztreonam	Amoxicillin
Total Number of Patients Entered	78	75
No. of Patients Excluded from the Efficacy Evaluation	22	35
Reasons:		
No pathogens or Resistant organisms isolated	14	20
Inappropriate Follow-up	6	9
Inappropriate duration of Therapy	0	3
Clinical diagnosis other than UTI	0	3
Other (No evaluable Patients in control group)	2	0
No. of Patients Evaluable for Efficacy**	56	40
Demographic Characteristics:		
Sex		
Female	53	39
Male	3	1
Age (Years)		
Range	19 - 82	19 - 79
Mean	34.8	38.3
Race		
Caucasian	51	34
Black	5	5
Not stated	0	1
No. of patients with symptomatic UTI	55	40
Duration of symptoms prior to therapy(days)	1 - 16	1 - 22
Mean (days)	3.8	4.6

* The investigators did not enter evaluable patients into both treatment groups.
** Patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy.

Protocol 18554-15: Comparison of Aztreonam (IM) vs. Amoxicillin (PO) in the Treatment of Acute, Uncomplicated Urinary Tract Infection.

Table II

Bacteriological Response*

	Aztreonam				Amoxicillin			
Pathogen	E	P	RL	RI	E	P	RL	RI
Escherichia coli	38/50	9	3	2(L)(a)	31/37	1	3	2(E)
Klebsiella pneumoniae	0/4	1	0	3(E)	1/1	0	0	0
Enterobacter aerogenes	1/1	0	0	0	0/0	-	-	-
Proteus mirabilis	0/0	-	-	-	2/2	0	0	0
Citrobacter sp.	1/1	0	0	0	0/0	-	-	-
Total	40/56 (71%)	10 (18%)	3 (5%)	5(3-E) (2-L) (9%)	34/40 (85%)	1 (2.5%)	3 (7.5%)	2(E) (5%)

* At 4-6 weeks after completion of therapy.
E - eradication of the original pathogen; number eradicated/number treated.
P - persistence of the original pathogen during and/or within 5-9 days post-therap
RL - relapse: re-emergence of the original pathogen at 4-6 weeks post-therapy.
RI - reinfection: emergence of a new pathogen(s) at post-therapy.
(L)- reinfection at 4-6 weeks post-therapy.
(E)- reinfection at 5-9 days post-therapy.

Bacteriologic Cure At 4-6 Weeks Post-therapy
(eradication of original pathogen)

Number Cured/Number Treated**
(cure rate)

Aztreonam	Cefamandole
40/53*(75.5%)	34/38*(89.5%)

**The patients with early reinfection (5-9 days post-therapy) who had no further follow-up were not included.

Protocol 18554-13: Comparison of Aztreonam (I.M.) vs. Cefamandole (I.M.) in the Treatment of Serious Gram-negative Urinary Tract Infection (UTI)

This is a multicenter, randomized, controlled study of aztreonam and cefamandole in the treatment of hospitalized patients with gram-negative urinary tract infections. Fourteen principal investigators, 7 each, domestic and foreign, participated in the study. The patients with clinical symptoms and signs compatible with urinary tract infections (UTIs) were assigned to either aztreonam or cefamandole according to the 2:1 randomization table. The dosage regimen of the two drugs was 1 g every 8 hours, intramuscularly for 5 to 10 days. The patient population in this study differs from that of the earlier study in that the majority of them had recurrent and/or complicated UTI.
Appropriate examinations of the urine, such as WBC and bacterial counts of the urine sediment, and urine cultures and susceptibility testing of clinical isolates were done before, during, and after completion of therapy. Routine hematological work-up and blood chemistries were also done prior to and after completion of therapy.

The applicant analysed the results of this multicenter study by each investigator as well as by pooling all investigators' data, irrespective of the location of the study (domestic or foreign). They excluded the patients who developed superinfection from the efficacy evaluation, but included them in the safety evaluation. For the evaluation of efficacy this reviewer excluded the data from the investigators who did not contribute at least one evaluable patient to each treatment group, and the patients who had no 4-6 weeks post-therapy follow-ups for reasons other than a failure to respond to therapy and discontinuance of therapy due to adverse reactions. The patients with superinfection were included in the efficacy evaluation. The data from 3 domestic and 7 foreign investigators (2 each from Greece and Egypt, 1 each from Australia, Belgium and Finland) were analysed. The numbers of the patients treated by each investigator varied and were small, as shown in Table I (A-J).

Of the total 194 patients (88 domestic and 106 foreign) entered into the study, 85 patients (45 domestic and 40 foreign) were excluded from the evaluation of efficacy by this reviewer. The major reasons for exclusions were inadequate post-therapy follow-ups and a negative culture (10^5CFU/ml) of the pretreatment urine specimens. A total of 109 patients (43 domestic and 66 foreign) were evaluable for the efficacy. The demographics of the evaluable patients are presented in Table I (A-J). The ratios of complicated vs. uncomplicated UTI, and initial episodes vs. recurrent episodes were comparable between the two treatment groups. Escherichia coli was the predominant pathogen in both treatment groups; 60% in the aztreonam group and 74% in the cefamandole group. The results of efficacy analyses by this reviewer are presented in Table II (A-F) and those by the applicant in the applicant's Table 5B (Vol. 3.2 pp. 2 375-376). The criteria for bacteriologic cure defined by the applicant and the reviewer differed, as stated in the review of the preceding UTI study (18554-15). The cure rates reported by the applicant, therefore, were higher than those stated by the reviewer. Analyses of the bacteriologic data by both applicant and this reviewer, nevertheless, indicated that a more favorable response was seen in the aztreonam treatment group.

The overall bacteriologic cure rates in the domestic and foreign studies of both complicated and uncomplicated UTIs pooled by the applicant were 88% (69/78) in the aztreonam treated patients and 74% (32/43) in the cefamandole treated patients at 5-9 days post-therapy. This reviewer's evaluation at 4-6 weeks post-therapy, however, showed that the bacteriologic cure rates were 76.7% (46/60) and 53.3% (16/30), respectively. The numbers of the patients evaluated by this reviewer were smaller since those patients who developed superinfection or reinfection during or at 5-9 days post-therapy, respectively and in whom no further follow-ups were made were excluded for the evaluation of bacteriologic cure, as shown in Table II (H). The pathogens causing recurrent UTI (relapse or reinfection) were all susceptible to the drugs except for 3 (2 in the aztreonam group, and 1 in the cefamandole group); 1 of 2 E. coli strains was resistant to the test drug, but the susceptibility of the other strain was unknown; 1 P. aeruginosa strain was resistant to the control drug. The incidences of relapse, reinfection, and superinfection were similar in the two treatment groups. The bacteriological and clinical responses seen in the domestic and foreign studies of complicated and uncomplicated UTI were comparable in the test and control groups, as presented in Table II (A-G). As expected, the bacteriologic cure rates for complicated UTI were lower than those of uncomplicated UTI in both treatment groups: 63% (12/19) vs. 83% (34/41) in the aztreonam group, and 40% (6/15) vs. 67% (10/15) in the cefamandol group, as shown in Tabale II (H). The relapse and reinfection rates were similar in the complicated and uncomplicated UTI, but superinfection rate was higher in the complicated UTI. The bacteriological cure rates for both complicated and uncomplicated UTI were lower in the cefamandole group. The lower cure rates seen in the study population are not unexpected since the majority of the patients had recurrent and/or complicated UTI. The eradication of the pathogens is more difficult in these conditions, and the recurrence (relapse or reinfection) of UTI is more likely to occur. Of all the patients received drug therapy, superinfection caused by resistant organisms occurred in 10 patients : seven (4 with complicated UTI and 3 with uncomplicated UTI) in the aztrenonam group and three (2 with complicated UTI and 1 with uncomplicated UTI) in the cefamandole group. Superinfections were due to Streptococcus fecalis (enterococci) resistant to aztreonam in 6 and Candida albicans in 1 of the aztreonam-treated patients, and Pseudomonas aeruginosa resistant to cefamandole in 3 cefamandole-treated patients. Two of the six patients with S. fecalis superinfection were treated with ampicillin. The clinical responses (resolution or improvement of symptoms and signs) observed were similar in both treatment groups, as shown in Table II (H).

Safety was assessed in all patients (194: 88 domestic and 106 foreign) treated with the test and control drugs. One hundred thirty patients received aztreonam and sixty received cefamandole for 2 to 19 days (5-7 days in the majority). Routine hematology, blood chemistry and urinalysis were performed prior to, during and after completion of therapy.

The adverse reactions possibly or probably attributed to the drugs were reported in 20% (26/130) of the aztreonam-treated patients and 27% (17/64) in the cefamandole-treated patients. The drugs were discontinued in the two patients of the cefamandole group and in one of the aztreonam group. One patient in the aztreonam group and two patients in the cefamandole group had more than one reaction. Death, which was not attributed to drug therapy, occurred in one of the aztreonam-treated patients. The adverse reactions reported were as follows:

Number of Patients Treated	Aztreonam 130	Cefamandole 64
Adverse Reactions		
Clinical:	17	14
Rash	0	2
Rash/pruritus	0	1
Rash/urticaria/pruritus	1	0
Nausea/headache/pruritus	0	1
Diarrhea	0	1
Cold sweat	1	0
Local reaction at injection site (pain/ or erythema/induration)	15	9
Laboratory abnormalities:	10	5
Elevated AST(SGOT)/ALT(SGPT)	7 (108)	3 (51)
Elevated alkaline phosphatase/LDH	0 (110)	1 (53)
Eosinophilia	2 (110)	2 (55)
Increased serum creatinine	1 (114)	0

Note: A few patients had more than one adverse reaction.
The numbers in parentheses represent the number of patients in whom the laboratory tests were done.

Table I (A)

Protocol 18554-13: Comparison of Aztreonam (I.M.) vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigator and Investigator Number: C.E.Cox, M.D.; 121

	Aztreonam	Cefamandole
Total Number of Patients Entered	20	10
No. of Patients Not Evaluable for Efficacy	10	2
Reasons:		
No pathogens isolated	1	0
Inappropriate Follow-up	6	1
5 days of therapy due to AR	0	1
Surgical procedures or concurrent antimicrobial therapy	0	1
No. of Patients Evaluable for Efficacy*	10	8
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	4	3
Male	6	5
Age (Years)		
Range	34 - 85	34 - 90
Mean	60.4	58.8
Race		
Caucasian	1	1
Black	9	7
Clinical Diagnosis		
UTI	7	5
Pyelonephritis	1	2
Cystitis	2	1
Complicated UTI	7	7
Uncomplicated UTI	3	1
Dosage Regimen (Route of Administration)	1 g q 8 h (IM)	1 g q 8 (IM)
Duration of Treatment (days)		
Range	5 - 19	2 - 9
Mean	8.2	6.1

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table I (B)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigator and Investigator Number: H.S. Soroff, M.D. ; 4701

	Aztreonam	Cefamandole
Total Number of Patients Entered	15	9
No. of Patients Not Evaluable for Efficacy	7	3
Reasons:		
No pathogens isolated or resistant pathogens	4	2
Inappropriate Follow-up	1	0
5 days of therapy due to AR	2	0
Concurrent antimicrobial therapy	0	1
No. of Patients Evaluable for Efficacy*	8	6
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	1	0
Male	7	6
Age (Years)		
Range	58 - 75	59 - 74
Mean	65.8	65.3
Race		
Caucasian	8	5
Black	0	0
Other	0	1
Clinical Diagnosis		
UTI	7	5
Cystitis	1	1
Complicated UTI	4	5
Uncomplicated UTI	4	1
Dosage Regimen	1 g q 8 h, IM	1 g q 8 h, IM
Duration of Treatment (days)		
Range	5 - 8	5 - 8
Mean	6.1	6.2

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table I (C)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigator and Investigator Number: P.A. Mackowiak, M. D. ; 5023

	Aztreonam	Cefamandole
Total Number of Patients Entered	10	5
No. of Patients Not Evaluable for Efficacy	5	2
Reasons:		
No pathogens isolated or resistant pathogens	2	2
Inappropriate Follow-up	2	0
Other protocol deviation	1	0
No. of Patients Evaluable for Efficacy*	5	3
Demographic Characteristics of Evaluable Patients:		-
Sex		
Female	0	0
Male	5	3
Age (Years)		
Range	39 - 88	56 - 78
Mean	63.2	64.0
Race		
Caucasian	4	3
Black	1	0
Clinical Diagnosis		
UTI	2	2
Pyelonephritis	2	1
Cystitis	1	0
Complicated UTI		1
Uncomplicated UTI	4	2
Dosage Regimen	1 g q 8 h, IM	1 g q 8 h, IM
Duration of Treatment (days)		
Range	5 - 8	5 - 6
Mean	5.6	5.3

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Table I (D)

Investigator and Investigator Number: A. Shaker, M.D. (Egypt), 4078

	Aztreonam	Cefamandole
Total Number of Patients Entered	3	2
No. of Patients Evaluable for Efficacy*	3	2
Demographic Characteristics:		
Sex		
Female	0	0
Male	3	2
Age (Years)		
Range	32 - 49	44 - 44
Race		
Caucasian	3	2
Clinical Diagnosis		
Pyelonephritis	2	1
Cystitis	1	1
Complicated UTI	0	2
Recurrent UTI	1	0
Duration of Treatment (days)		
Range	5 - 5	5 - 7

* Patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy.

Table I (E)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator and Investigator Number: F.Y. Fyhrquist, M.D. (Finland), 5303

	Aztreonam	Cefamandole
Total Number of Patients Entered	9	2
No. of Patients Not Evaluable for Efficacy	5	0
Reasons:		
No pathogens isolated/pre-treatment cultures 48 hrs.	4	0
Inappropriate Follow-up	1	0
No. of Patients Evaluable for Efficacy*	4	2
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	4	2
Male	0	0
Age (Years)		
Range	30 - 55	67 - 68
Mean	43.5	67.5
Race		
Caucasian	4	2
Clinical Diagnosis		
Pyelonephritis	4	2
Complicated UTI	1	0
Uncomplicated UTI	3	2
Dosage Regimen	1 g q 8 h (IM)	1 g q 8 h (IM)
Duration of Treatment (days)		
Range	5 - 6	5 - 5
Mean	5.3	5

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table I (F)

Protocol 18554-13. Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator and Investigator Number: G. Daikos (Greece); 6310

	Aztreonam	Cefamandole
Total Number of Patients Entered	20	10
No. of Patients Not Evaluable for Efficacy	11	7
Reasons:		
No pathogens isolated	6	0
Inappropriate Follow-up	5	7
No. of Patients Evaluable for Efficacy*	9	3
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	4	3
Male	5	0
Age (Years)		
Range	27 - 76	34 - 56
Mean	54.1	45.3
Race		
Caucasian	9	3
Clinical Diagnosis		
Pyelonephritis	7	3
Cystitis	2	0
Complicated UTI	6	3
Uncomplicated UTI	3	0
Dosage Regimen	1 g q 8 h (IM)	1 g q 8 h (IM)
Duration of Treatment (days)		
Range	7 - 11	11
Mean	9.1	11

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table I (G)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator and Investigator Number: M. Tsouroutsoglu (Greece); 6312

	Aztreonam	Cefamandole
Total Number of Patients Entered	21	10
No. of Patients Not Evaluable for Efficacy	4	3
Reasons:		
Inappropriate Follow-up	4	3
No. of Patients Evaluable for Efficacy*	17	7
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	12	5
Male	5	2
Age (Years)		
Range	18 - 73	30 - 72
Mean	49.4	63.4
Race		
Caucasian	17	7
Clinical Diagnosis		
UTI (lower)	7	4
Pyelonephritis	10	3
Complicated UTI	2	2
Uncomplicated UTI	15	5
Dosage Regimen	1 g q 8 h (IM)	1 g q 8 h (IM)
Duration of Treatment (days)		
Range	5 - 10	6 - 10
Mean	6.7	7.4

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table I (H)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator and Investigator Number: L M. Verbist, M.D.(Belgium) , 6346

	Aztreonam	Cefamandole
Total Number of Patients Entered	9	5
No. of Patients Not Evaluable for Efficacy	3	1
Reasons:		
No pathogens isolated/cultures 48 h pretherapy	2	1
Inappropriate Follow-up	1	0
No. of Patients Evaluable for Efficacy*	6	4
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	6	3
Male	0	1
Age (Years)		
Range	54 - 82	45 - 82
Mean	69.8	63.3
Race		
Caucasian	6	4
Clinical Diagnosis		
Cystitis (uncomplicated)	6	4
Dosage Regimen	1 g q 8 h (IM)	1 g q 8 h (IM)
Duration of Treatment (days)		
Range	5 - 5	5 - 5
Mean	5	5

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table I (I)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator and Investigator Number: M. Sabbour, M.D. (Egypt); 7539

	Aztreonam	Cefamandole
Total Number of Patients Entered	4	3
No. of Patients Not Evaluable for Efficacy	2	2
Reasons:		
No pathogens isolated/colony count not recorded	2	2
No. of Patients Evaluable for Efficacy*	2	1
Demographic Characteristics:		
Sex		
Female	0	1
Male	2	0
Age (Years)		
Range	42 - 53	28
Race		
Caucasian	2	1
Clinical Diagnosis		
Pyelonephritis	2	1
Complicated UTI	1	1
Uncomplicated UTI	1	0
Duration of Treatment (days)		
Range	7 - 7	6

* Patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy.

Table I (J)

Protocol 18354-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator and Investigator Number: G.E. Rich, M.D. (Australia); 7570

	Aztreonam	Cefamandole
Total Number of Patients Entered	6	2
No. of Patients Not Evaluable for Efficacy	2	0
Reasons:		
Inappropriate Follow-up	1	0
5 days of therapy	1	0
No. of Patients Evaluable for Efficacy*	4	2
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	4	2
Male	0	0
Age (Years)		
Range	35 - 75	70 - 72
Mean	63.3	71.5
Race		
Caucasian	4	2
Clinical Diagnosis		
UTI	3	2
Cystitis	1	0
Uncomplicated UTI	4	2
Dosage Regimen	1 g q 8 h (IM)	1 g q 8 h (IM)
Duration of Treatment (days)		
Range	5 - 5	5 - 5
Mean	5	5

* Include patients who had appropriate follow-ups up to 4-6 weeks after completion of therapy, and those with superinfection.

Table II (A)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 0121; 4701; 5203

Complicated Urinary Tract Infection

Bacteriologic Response*

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
E. coli	2/4	1	1	1(L)	0	3/6	2	1	1(L)	0
P. mirabilis	1/1	0	0	0	0	0/0	-	-	-	-
P. vulgaris	1/1	0	0	0	0	0/1	0	0	1(E)	0
K. pneumoniae	1/3	1	0	0	1	1/1	0	0	0	0
E. cloacae	0/0	-	-	-	-	0/1	0	0	0	1
P. aeruginosa	0/1	0	0	0	1	0/0	-	-	-	-
E. coli + K. pneumoniae	1/1	0	0	0	0	0/0	-	-	-	-
E. coli + P. mirabilis	0/1	0	0	0	1	0/0	-	-	-	-
P. mirabilis + P. stuartii	0/0	-	-	-	-	0/1	1**	0	0	1**
Total	6/12	2	1	1(L)	3	4/10	3	1	2 (1-E) (1-L)	2

* At 4-6 weeks after completion of therapy
** The same patient
E - eradicated: Number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: Emergence of new pathogen resistant to the study drug during treatment

Table II (B)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 0121; 4701; 5203

Uncomplicated Urinary Tract Infection

Bacteriologic Response*

Pathogen	Aztreonam					Cefamandole				
	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	4/4	0	0	0	0	1/2	0	1	0	0
P. mirabilis	0/0	-	-	-	-	0/1	0	0	1(E)	0
K. pneumoniae	1/1	0	0	0	0	1/3	2	0	0	0
K. oxytoca	1/1	0	0	1(L)	0	0/0	-	-	-	-
E. cloacae	2/2	0	0	1(L)	0	0/0	-	-	-	-
P. stuartii	1/1**	0	0	1**(E)	0	0/0	-	-	-	-
P. aeruginosa	2/2	0	0	0	0	0/0	-	-	-	-
Total	11/11	0	0	3(2-L)	0	2/6	2	1	1(E)	0

* At 4-6 weeks after completion of therapy
** The same patient.
E - eradicated: Number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: Emergence of new pathogen resistant to the study drug during treatment

Table II (C)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 0121; 4701; 5203

Urinary Tract Infections (Complicated and Uncomplicated)

Bacteriologic Response*

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
E. coli	6/8	1	1	1(L)	0	4/8	2	2	1(L)	0
K. pneumoniae	2/4	1	0	0	1	2/4	2	0	0	0
P. mirabilis	1/1	0	0	0	0	0/1	0	0	1(E)	0
P. vulgaris	1/1	0	0	0	0	0/1	0	0	1(E)	0
K. oxytoca	1/1	0	0	1(L)	0	0/0	-	-	-	-
E. cloacae	2/2	0	0	1(L)	0	0/1	0	0	0	1
P. aeruginosa	2/3	0	0	0	1	0/0	-	-	-	-
P. stuartii	1/1**	0	0	1**(E)	0	0/0	-	-	-	-
E. coli + K. pneumoniae	1/1	0	0	0	0	0/0	-	-	-	-
E. coli + P. mirabilis	0/1	0	0	0	1	0/0	-	-	-	-
P. mirabilis + P. stuartii	0/0	-	-	-	-	0/1	1**	-	-	1**
Total	17/23	2	1	4(3-L) (1-E)	3	6/16	5	2	3(1-L) (2-E)	2

* At 4-6 weeks after completion of therapy
** The same patient
E - eradicated: Number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: Emergence of new pathogen resistant to the study drug during treatment

Table II (D)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 0121; 4701; 5203

Urinary Tract Infections (Complicated and Uncomplicated)

Bacteriologic Cure at 4-6 Weeks Post-therapy
(eradication of original pathogen)

Number Cured/Number Treated*
(cure rate)

Aztreonam	Cefamandole
17/20 (85%)	6/13 (46%)

Clinical Cure/Improvement at 4-6 Weeks Post-therapy

Number Cured + Improved/Number Treated*

Aztreonam	Cefamandole
20/20 (100%)	11/13 (84.6%)

*Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy who had no further follow-ups were not included.

Table II (E)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator Number: 4078, 5303; 6310, 6312; 6346, 7539; 7570

Complicated Urinary Tract Infection

Bacteriologic Response*

	Aztreonam					Cefamandole				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	3/6	1	1	0	1	2/7	3	1	1(E)	0
P. mirabilis	2/3	0	0	1(E)	1	0/1	1	0	0	0
K. pneumoniae	0/1	0	1	0	0	0/0	-	-	-	-
Citrobacter sp.	0/1	0	0	1(E)	0	0/0	-	-	-	-
P. aeruginosa	1/2	1	0	0	0	0/0	-	-	-	-
Total	6/13 (46%)	2	2	2(E)	1	2/8 (25%)	4	1	1(E)	0

* At 4-6 weeks after completion of therapy
E - eradicated: Number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: Emergence of new pathogen resistant to the study drug during treatment

Table II (F)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator Number: 4078, 5303; 6310; 6312; 6346; 7539, 7570

Uncomplicated Urinary Tract Infection

Bacteriologic Response*

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
E. coli	17/24	4	2	3 (2-L) (1-E)	0	7/10	1	0	3 (2-L) (1-E)	1
P. mirabilis	5/6	0	1	1(L)	0	0/1	1	0	0	0
P. fluorescense	1/1	0	0	0	0	0/0	-	-	-	-
E. coli + K. pneumoniae	0/0	-	-	-	-	1/1	0	0	0	0
Total	23/31 (74%)	4	3	4 (3-L) (1-E)	0	8/12 (67%)	2	0	3 (2-L) (1-E)	1

* At 4-6 weeks after completion of therapy
E - eradicated: Number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: Emergence of new pathogen resistant to the study drug during treatment

Table II (G-1)

Protocol 18554-13: Comparison of Aztreonam vs. Cefama[illegible] the Treatment of Serious Gram-negative Urinary Trac[illegible]ction

Foreign Study

Urinary Tract Infections (Complicated and Uncomplicated)

Bacteriologic Response*

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
E. coli	20/30	5	3	3 (1-E) (2-L)	1	9/17	4	1	4 (2-E)	1
P. mirabilis	7/9	0	1	2 (1-E) (1-L)	0	0/0	2	0	0	0
K. pneumoniae	0/1	0	1	0	0	0/0	-	-	-	-
C. freundii	0/1	0	0	1(E)	0	0/0	-	-	-	-
P. aeruginosa	1/2	1	0	0	0	0/0	-	-	-	-
P. fluorescens	1/1	0	0	0	0	0/0	-	-	-	-
E. coli + K. pneumoniae	0/0	-	-	-	-	1/1	-	-	-	0
Total	29/44 (66%)	6	5	6 (3-E) (3-L)	1	10/20 (50%)	6	1	4 (2-E) (2-L)	1

* At 4-6 weeks after completion of therapy
E - eradicated: Number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: Emergence of new pathogen resistant to the study drug during treatment

Table II (G-2)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Urinary Tract Infections (Complicated and Uncomplicated)

Bacteriologic Cure at 4-6 Weeks Post-therapy
(eradication of original pathogen)

Number Cured/Number Treated* (cure rate)	
Aztreonam	Cefamandole
29/40 (72.5%)	10/17 (58.8%)

Clinical Cure/Improvement At 4-6 Weeks Post-therapy

Number Cured + Improved/Number Treated*	
Aztreonam	Cefamandole
37/40 (92.5%)	16/17 (94%)

*Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Table II (H)

Protocol 18554-13: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic and Foreign Studies Pooled:

Bacteriologic Cure at 4-6 Weeks Post-therapy
(eradication of original pathogen)

UTI	Number Cured/Number Treated* (cure rate)	
	Aztreonam	Cefamandole
Complicated	12/19 (63.2%)	6/15 (40%)
Uncomplicated	34/41 (82.9%)	12/17 (66.7%)
Total	46/60 (76.7%)	16/30 (53.3%)

Clinical Cure/Improvement At 4-6 Weeks Post-therapy*

UTI	Number Cured + Improved/Number Treated	
	Aztreonam	Cefamandole
Complicated	18/19 (94.7%)	13/15 (86.7%)
Uncomplicated	39/41 (95%)	14/15 (93%)
Total	57/60 (95%)	27/30 (90%)

*Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Protocol 18554-14: Comparison of Aztreonam (I V.) vs. Cefamandole (I.V.) in the Treatment of Serious Gram-negative Urinary Tract Infection

As in the preceding study (Protocol 18553-13), the efficacy and safety of aztreonam were compared to those of cefamandole in the treatment of serious urinary tract infections caused by aerobic gram-negative pathogens in hospitalized patints. According to a randomization schedule, the patients were allocated to receive either aztreonam or cefamandole at a ratio of 2:1, respectively. One foreign investigator did not adhere to the randomization scheme.
The criteria for entrance to, and exclusion from the study were similar to those of the preceding study, as were the procedures for monitoring the patients. The dosage and duration of therapy were similar to those in the preceding UTI study, except that the drugs were administered intravenously rather than intramuscularly.

A total of 13 principal investigators (9 domestic and 4 foreign) who participated in this study enrolled a total of 320 patients (269 domestic and 51 foreign). Two domestic investigators (0121 and 6208) treated more than one half of all patients entered into this study. Two-hundred-thirteen patients were treated with aztreonam, 105 patients with cefamandole, and the remaining 2 with no drug. One-hundred-eight (51%) of the 213 patients in the aztreonam group and 51 (49%)of the 105 patients in the cefamandole group were evaluable for efficacy. Of the 159 evaluable patients, 151 patients were treated by domestic investigators, and the remaining 8 patients by foreign investigators.

The demography of the evaluable patients in the two treatment groups was similar, as shown in the reviewer's Table I (A-D). The majority of the patients had complicated (intrinsic or extrinsic urinary tract abnormalities) and/or recurrent UTI. Upper urinary tract infection (pyelonephritis) was the predominant clinical diagnosis recorded by the investigators in this study. Localization studies, however, were not performed. The most common urinary pathogens isolated were Escherichia coli (62%) followed by Klebsiella pneumoniae (1.6%), as presented in Table II (C). The reasons for exclusion of patients from the efficacy evaluation are listed in Table I (A-D).

Bacteriological responses observed in this study population are presented in Table II (A-C and E). In the domestic study, the overall bacteriological and clinical responses seen in both complicated and uncomplicated UTI were more favorable in the aztreonam group, as shown in Table II (D). The bacteriologic cure rates for complicated UTI were 76% (32/42) and 56% (10/18) in the aztreonam-treated patients and cefamandole-treated patients, respectively. For uncomplicated UTI, the cure rates were 81% (30/37) and 58% (11/19), respectively. The combined (complicated + uncomplicated) cure rates were 78.5% (62/79) in the aztreonam treatment group and 56.8% (21/37) in the cefamandole treatment group. The cure rates for the most common uropathogen, E. coli, were 79.5% (39/49) in the aztreonam group and 52.2% (12/23) in the cefamandole group. The number of the patients treated in the foreign study is very small, as shown in Table II (E-F). The rates of relapse (re-emergence of the original pathogen 10^5CFU/ml at 4-6 weeks post-therapy) and superinfection (emergence of resistant pathogen 10^5CFU/ml during therapy) were similar in the two treatment groups, but the rate of reinfection (emergence of a new pathogen 10^5CFU/ml) was higher in the cefamandole treatment group (26.1%), as compared to the aztreonam group (15.7%).

Pseudomonas aeruginosa in the cefamandole-treated patients. A few of these patients were treated with other antibiotics.
The overall microbiologic cure rates reported by the applicant were 87% (116/134) in the aztreonam group and 76% (41/54) in the cefamandole group. The clinical cure rates were 99% (133/134) and 100% (54/54) in the aztreonam group and the cefamandole group, respectively. The differences in the numbers of evaluable patients and cure rates reported by the applicant and this reviewer are primarily attributable to this reviewer's exclusion of those patients who were considered as 'cured' by the applicant at 5-9 days post-therapy, but who had no further follow-up at 4-6 weeks, and to the reviewer's consideration of those patients with "late" relapse (at 4-6 weeks post-therapy) as failures. The analyses of the bacteriologic data nevertheless indicate that aztreonam appears to be more effective than cefamandole, a marketed second generation cephalosporin, in the treatment of complicated and/or recurrent UTI caused by gram-negative uropathogens.

The Safety of the drugs was evaluated in all patients who received the test and the control drugs. The clinical adverse reactions possibly or probably related to the drug therapy occurred in 19 (8.9%) of the 213 patients treated with aztreonam and 1 (0.95%) of the 105 patients treated with cefamandole. Phlebitis/thrombophlebitis was the leading adverse reaction associated with aztreonam in this study. Laboratory abnormalities, primarily increases in transaminase (ALT/AST) levels, were observed in 25 (13.5%) of the 185 aztreonam-treated patients and 4 (4.7%) of the 86 cefamandole-treated patients. In 8 of the 25 patients, the transaminase levels were greater than 100 IU/ml, but the increases were transient, and no clinical manifestations of hepatotoxicity were observed. Five deaths occurred, 4 in the aztreonam and 1 in the cefamandole group. None of the deaths were attributed to drug therapy. The adverse reactions observed were as follows:

	Aztreonam	Cefamandole
No. of Patients Treated	213	105
Adverse Reactions:		
Clinical:	19	1
Nausea	1	0
Diarrhea	1	1
Rash	2	0
Pruritus	1	0
Headache	1	0
Phlebitis	8	0
Pain at injection (IM) site	1	0
Laboratory abnormalities::		
Eosinophilia	1 (185)	0
ALT(SGPT)/AST(SGOT)	25 (185)	4 (86)
Alkaline phosphatase	2 (185)	0
LDH	2 (175)	0

The numbers in parentheses represent the number of patients in whom the laboratory tests were done.

Conclusions: The results of this multicenter, randomized, controlled study of aztreonam indicated that aztreonam was more efficacious than the control drug, cefamandole, a second generation cephalosporin, in the treatment of hospitalized patients with complicated and uncomplicated urinary tract infections caused by aerobic gram-negative uropathogens, particularly E. coli. The safety of aztreonam, however, was less favorable than cefamandole.

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The overall microbiologic cure rates reported by the applicant were 87% (116/134) in the aztreonam group and 76% (41/54) in the cefamandole group. The clinical cure rates were 99% (133/134) and 100% (54/54) in the aztreonam group and the cefamandole group, respectively. The differences in the numbers of evaluable patients and cure rates reported by the applicant and this reviewer are primarily attributable to this reviewer's exclusion of those patients who were considered as 'cured' by the applicant at 5-9 days post-therapy, but who had no further follow-up at 4-6 weeks, and to the reviewer's consideration of those patients with "late" relapse (at 4-6 weeks post-therapy) as failures. The analyses of the bacteriologic data nevertheless indicate that aztreonam appears to be more effective than cefamandole, a marketed second generation cephalosporin, in the treatment of complicated and/or recurrent UTI caused by gram-negative uropathogens.

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Diarrhea	1	1
Rash	2	0
Pruritus	1	0
Headache	1	0
Phlebitis	8	0
Pain at injection (IM) site	1	0
Laboratory abnormalities::		
Eosinophilia	1 (185)	0
ALT(SGPT)/AST(SGOT)	25 (185)	4 (86)
Alkaline phosphatase	2 (185)	0
LDH	2 (175)	0

The numbers in parentheses represent the number of patients in whom the laboratory tests were done.

Conclusions: The results of this multicenter, randomized, controlled study of aztreonam indicated that aztreonam was more efficacious than the control drug, cefamandole, a second generation cephalosporin, in the treatment of hospitalized patients with complicated and uncomplicated urinary tract infections caused by aerobic gram-negative uropathogens, particularly E. coli. The safety of aztreonam, however, was less favorable than cefamandole.

Table I (A)

Protocol 18554-14: Comparison of Aztreonam (I V.) vs. Cefamandole (I.V.) in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigator and Investigator Number: S.J. Childs, M.D. ; 6208

	Aztreonam	Cefamandole
Total Number of Patients Entered	74	37
No. of Patients Not Evaluable for Efficacy	20	14
Reasons:		
No pathogens isolated/or recorded	2	1
Resistant organisms	0	6
Inappropriate Follow-up	9	2
5 days of therapy	1	0
Surgical procedure or concurrent antimicrobial therapy	1	1
Clinical diagnoses other than UTI (prostatitis; epididymitis)	7	5
No. of Patients Evaluable for Efficacy*	54	23
Demographic Characteristics:		
Sex		
Female	45	20
Male	19	3
Age (Years)		
Range	19 - 87	28 - 58
Mean	57	53
Race		
Caucasian	49	21
Black	5	2
Clinical Diagnosis		
Pyelonephritis	36	12
Cystitis	18	11
Complicated/Recurrent UTI	28	11
Uncomplicated UTI	26	12
Dosage Regimen	1-2 g q 8-12 h	1-2 g q 8-12 h
Total Dose (Range)	6 - 37 g	15 - 39 g
Duration of Treatment (days)		
Range	5 - 11	5 - 9
Mean	7.1	6.6

* Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table I (B)

Protocol 18554-14: Comparison of Aztreonam (I V.) vs. Cefamandole (I.V.) in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigator and Investigator Number: C.E. Cox, M.D. ; 0121

	Aztreonam	Cefamandole
Total Number of Patients Entered	33	16
No. of Patients Not Evaluable for Efficacy	11	6
Reasons:		
No pathogens isolated/or recorded	1	0
Inappropriate Follow-up	8	6
5 days of therapy due to AR	1	0
Surgical procedure	1	0
No. of Patients Evaluable for Efficacy*	22	10
Demographic Characteristics:		
Sex		
Female	4	4
Male	18	6
Age (Years)		
Range	25 - 86	25 - 85
Mean	65	62
Race		
Caucasian	7	
Black	15	
Clinical Diagnosis		
UTI (unspecified)	19	9
Pyelonephritis	2	1
Cystitis	1	0
Complicated/Recurrent UTI	20	7
Uncomplicated UTI	2	3
Dosage Regimen	1-2 g q 8-12 h	1-2 g q 8-12 h
Total Dose (Range)	6 - 37 g	15 - 39 g
Duration of Treatment (days)		
Range	5 - 11	5 - 11
Mean	7.7	7.2

* Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table I (C)

Protocol 18554-14: Comparison of Aztreonam (I V.) vs. Cefamandole (I.V.) in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

No. of Principal Investigators: 6

Investigators' Numbers: Soroff (4701); Apicella (6207), Gentry (6210); Johnson (6213); Wyle (6215); Farrar (6265).

	Aztreonam	Cefamandole
Total Number of Patients Entered	68	34
No. of Patients Not Evaluable for Efficacy	42	18
Reasons:		
No pathogens isolated/or recorded	9	3
Resistant microorganism	3	3
Inappropriate Follow-up	23	11
5 days of therapy due to AR or other cause	2	0
Surgical procedure or concurrent antimicrobial therapy	4	0
Death	1	1
No. of Patients Evaluable for Efficacy*	26	16
Demographic Characteristics:		
Sex		
Female	17	9
Male	9	7
Age (Years)		
Range	23 - 80	26 - 87
Mean	52	56
Race		
Caucasian	18	10
Black	8	6
Clinical Diagnosis		
Pyelonephritis	15	10
Cystitis/Lower UTI	10	5
UTI (unspecified)	1	1
Complicated/Recurrent UTI	10	6
Uncomplicated UTI	16	10
Dosage Regimen	1-2 g q 8 h	1-2 g q 6 -8 h
Duration of Treatment (days)		
Range	5 - 13	5 - 16
Mean	7.3	8.8

* Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table I (D)

Protocol 18554-14: Comparison of Aztreonam (I.V.) vs. Cefamandole (I.V.) in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

No. of Principal Investigators: 3

Investigators' Numbers: Yourassowsky (6296; Belgium); Wurth(6364, Netherland), Westenfelder (6268; West Germany)**.

	Aztreonam	Cefamandole
Total Number of Patients Entered	33	17
No. of Patients Not Evaluable for Efficacy	27	15
Reasons:		
No pathogens isolated	1	0
Inappropriate follow-up	2	4
5 days of therapy	1	1
Concurrent antimicrobial therapy	1	0
Clinical diagnosis other than UTI	1	0
Route of Administration (I.M.)	1	0
Randomization not done**	20	10
No. of Patients Evaluable for Efficacy*	5	2
Demographic Characteristics:		
Sex		
Female	2	2
Male	4	0
Age (Years)		
Range	19 - 79	32 - 57
Mean	53	45
Race		
Caucasian	6	0
Not recorded	0	1
Clinical Diagnosis		
Pyelonephritis	4	2
Cystitis/Lower UTI	1	0
UTI (unspecified)	1	0
Complicated/Recurrent UTI	6	1
Uncomplicated UTI	0	1
Dosage Regimen	1 g q 8 h	0.5 - 1 g q 8 h
Duration of Treatment (days)		
Range	6 - 14	7 - 9
Mean	8	7

* Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table II (A)

Protocol 18554-14: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 0121; 4701; 6207, 6208, 6210, 6213; 6215.

Complicated Urinary Tract Infection
Bacteriologic Response*

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
E. coli	16/32	6	0	4(3-E)	8(a)	4/14	4	0	7(E)	1
K. pneumoniae	8/10	1	0	1(L)	1	0/0	-	-	-	-
P. mirabilis	2/4	0	1	0	1	1/2	1**	0	0	1**
P. vulgaris	1/1	0	0	0	0	1/1**	0	0	1(E)**	0
P. aeruginosa	0/1	1	0	0	0	0/0	-	-	-	-
P. rettgeri	1/1	0	0	0	0	0/0	-	-	-	-
K. oxytoca	1/1	0	0	1(L)	0	0/0	-	-	-	-
E. aerogenes	0/0	-	-	-	-	2/2	0	0	0	0
E. cloacae	1/1	1	0	1(E)**	1**	2/2	0	0	0	0
C diversus	0/0	-	-	-	-	0/1	1	0	0	0
C. freundii	1/2	0	0	1(E)**	1**	0/0	-	-	-	-
M. morganii	0/0	-	-	-	-	0/1	1	0	0	0
S. marcescens	1/1	0	0	0	0	0/0	-	-	-	-
E. coli + K. pneumoniae	0/1	0	0	1(E)	0	0/1	1	0	0	0
K. pneumoniae + P. mirabilis + C. diversus	0/0	0	0	0	1	0/0	-	-	-	-
Total	32/58 (55%)	9	1	9(6-E) (3-L)	13	10/24 (42%)	8	0	8(E)	2

*At 4-6 weeks after completion of therapy. ** The same patient.
E - eradication; number eradicated/number treated.
P - persistence ; RL - relapse ; RI - reinfection
SI - superinfection: (a) in 2 of 8 SI, the original pathogen was eradicated
(E) - reinfection at 5-9 days after completion of therapy.
(L) - reinfection at 4-6 weeks after completion of therapy.

Table II (B)

Protocol 18554-14: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Uncomplicated Urinary Tract Infection
Bacteriologic Response *

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
E. coli	23/31	4	0	5(4-E)	0	8/17	6	1	1(E)	2(a)
K. pneumoniae	4/6	0	1	1(E)	1(b)	1/4	1	0	2(1-E)	1
P. mirabilis	1/1	0	0	0	0	1/1	0	0	1(L)	0
P. aeruginosa	1/2	0	1	0	2(c)	0/0	-	-	-	0
K. oxytoca	0/0	-	-	-	-	0/1	0	0	1(E)	0
E. aerogenes	0/1	1	0	0	0	0/0	-	-	-	-
C. freundii	0/1	0	0	1(E)	0	1/1	0	0	0	0
C. diversus	1/1	0	0	0	0	0/0	-	-	-	0
E. coli + P. mirabilis	0/1	0	0	0	1	0/0	-	-	-	0
E. coli+ M. morganii	0/0	-	-	-	0	0/1	0	0	0	1
Total	30/44 (68%)	5	2	7(6-E) (1-L)	4	11/25 (44%)	7	1	5(3-E) (2-L)	4

* At 4-6 weeks after completion of therapy ** The same patients
E- eradication; number eradicated/number treated
P - persistence
RL - relapse
RI - reinfection
SI - superinfection: (a) The original pathogen persisted (P) in one
(b) The original pathogen was eradicated (E) in one
(c) The original pathogen was eradicated (E) in one, and re-emerged (RI) in the other
(E) - Reinfection at 5-9 days after completion of therapy
(L) - Reinfection at 4-6 weeks after completion of therapy

Table II (C)

Protocol 18554-14: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 0121; 4701; 6207; 6208, 6210, 6213; 6215; 6265

Urinary Tract Infections (Complicated and Uncomplicated)
Bacteriologic Response*

Pathogen	Aztreonam E	P	RL	RI	SI	Cefamandole E	P	RL	RI	SI
Single Pathogen:										
E. coli	39/63	10	0	9(7-E)	8(a)	12/31	10	1	8(E)	3(b)
K. pneumoniae	12/16	1	1	2(1-E)	2(c)	1/4	1	0	2(1-E)	1
P. mirabilis	3/5	0	1	0	1	2/3	1**	0	1(L)	1**
P. vulgaris	1/1	0	0	0	0	1/1**	0	0	1(E)**	0
P. aeruginosa	1/3	1	1	0	2(d)	0/0	-	-	-	-
P. rettgeri	1/1	0	0	0	0	0/0	-	-	-	-
K. oxytoca	1/1**	0	0	1(L)**	0	0/1	-	-	1(E)	0
E. aerogenes	0/1	1	0	0	0	2/2	0	0	0	0
E. cloacae	1/3	1	0	1(E)**	1**	2/2	0	0	0	0
C diversus	1/1	0	0	0	0	0/1	1	0	0	0
C. freundii	1/3	0	0	2(E)	1	1/1	0	0	0	0
M. morganii	0/0	-	-	-	-	0/1	1	0	0	0
S. marcescens	1/1	0	0	0	0	0/0	-	-	-	-
Multiple Pathogens:										
E. coli + K. pneumoniae	1/1	0	0	1(E)	0	0/1	1	0	0	0
E. coli + P. mirabilis	0/1	0	0	0	1	0/0	-	-	-	-
E. coli + M. morganii	0/0	-	-	-	-	0/1	0	0	0	1
K. pneumoniae + P. mirabilis + C. diversus	0/1	0	0	0	1	0/0	-	-	-	-
Total	62/102 (59.6%)	14	3	16 (12-E) (4-L)	17	21/49 (42.9%)	15	1	13 (11-E) (2-L)	6

* At 4-6 weeks after completion of therapy ** The same patients
E - eradication; number eradicated/number treated
P - persistence ; RL - relapse ; RI - reinfection
SI - superinfection: (a) The original pathogen was eradicated (E) in 2 of 8 SI
(b) The original pathogen was eradicated (E) in 1 of 3 SI
(d) The original pathogen was eradicated (E) in 1 of 2 SI
(d) The original pathogen was eradicated (E) in 1, and re-emerged (RI) in 1
(E) - reinfection at 5-9 days after completion of therapy (post-therapy)
(L) - reinfection at 4-6 weeks after completion of therapy (post-therapy)

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Table II (D)

Protocol 18554-14: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Bacteriologic Cure at 4-6 Weeks Post-therapy
(eradication of original pathogen)

UTI	Number Cured/Number Treated* (cure rate)	
	Aztreonam	Cefamandole
Complicated	32/42 (76%)	10/18 (55.5%)
Uncomplicated	30/37 (81.1%)	11/19 (57.9%)
Total	62/79 (78.5%)	21/37 (56.8%)

Clinical Cure/Improvement At 4-6 Weeks Post-therapy

UTI	Number Cured + Improved/Number Treated	
	Aztreonam	Cefamandole
Complicated	38/42 (90.5%)	9/18 (50%)
Uncomplicated	32/36 (88.9%)	14/19 (73.7%)
Total	70/78 (89.7%)	23/37 (62.2%)

*Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Table II (E)

Protocol 18554-14: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator Number: 6296, 6364.

Complicated Urinary Tract Infection
Bacteriologic Response*

	Aztreonam					Cefamandole				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	0/2	0	1	0	1	1/1	0	0	0	0
P. mirabilis	0/1	1	0	0	0	0/C	-	-	-	-
P. aeruginosa	1/2	0	1	1(E)(a)	0	0/0	-	-	-	-
S. marcescens	1/1	0	0	0	0	0/0	-	-	-	-
Total	2/6	1	2	1(E)	1	1/1	0	0	0	0

Uncomplicated Urinary Tract Infection
Bacteriologic Response*

	Aztreonam					Cefamandole				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	0/0	-	-	-	-	1/1	0	0	0	0

* At 4-6 weeks after completion of therapy ** The same patients
E- eradication, number eradicated/number treated
P - persistence
RL - relapse
RI - reinfection the original pathogen was eradicated
SI - superinfection
E - reinfection at 5-9 days post-therapy; (a)the original pathogen was eradicated.

Table II (F)

Protocol 18554-14: Comparison of Aztreonam vs. Cefamandole in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Investigator Number: 6296; 6364.

Bacteriologic Cure at 4-6 Weeks Post-therapy
(eradication of original pathogen)

UTI	Number Cured/Number treated* (cure rate)	
	Aztreonam	Cefamandole
Complicated	2/6	1/1
Uncomplicated	0/0	1/1
Total	2/6	2/2

Clinical Cure/Improvement At 4-6 Weeks Post-therapy

UTI	Number Cured + Improved/Number Treated*	
	Aztreonam	Cefamandole
Complicated	4/5	1/1
Uncomplicated	0/0	1/1
Total	4/5	2/2

*Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Urinary Tract Infection

This was a multicenter, randomized, controlled study of aztreonam and an aminoglycoside (gentamicin or netilmicin) in hospitalized patients with aerobic gram-negative urinary tract infections. The randomization of the patients to the test and the control drug, were made in a 2: 1 ratio, respectively, as in the preceding UTI study. The dosage regimens were: aztreonam - 1-2 g. q 8 h; gentamicin - 1 mg/kg q 8 h, intravenously or intramuscularly, and netilmicin, 150 mg q 12 h intravenously. The majority of patients received the drugs intravenously. The duration of therapy ranged from 5 to 16 days, with a mean duration of 8.7 days in the aztreonam group, and ranged from 5 to 10 days, with a mean of 7.6 days in the control group, as shown in Table I(A). In the foreign studies, a few patients were treated with netilmicin, as shown in Table II (B). The methods of this multicenter study were similar to those of the preceding multiple dose UTI studies except for the control drug used.

Eleven principal investigators (4 domestic and 7 foreign) enrolled a total of 127 patients (67 domestic and 59 foreign) in this study. Eighty-six patients were treated with aztreonam, thirty-five with gentamicin, and five with netilmicin. The demographic characteristics of the treatment groups were similar, with respect to age, weight, and race, as shown in the applicant's Tables 2A and 2 B (vol. 3.2: 563-4). Forty (46.5%) of the aztreonam-treated patients and 19 (55.8%) of the gentamicin-treated patients, and 1 (20%) of the netilmicin-treated patients were not evaluable for efficacy, for reasons listed in Table I (A and B). The demographics of the evaluable patients in the efficacy analyses by this reviewer were also similar in both treatment groups, as presented in Table I (A and B). In this study, cystitis was the clinical diagnosis in the majority of cases, 71.1% of the aztreonam group and 98.8% of the aminoglycosides group. However, sixty percent of the aztreonam-treated patients and 47% of the aminoglycosides-treated patients had complicated UTI.

The methods of evaluation of efficacy by the applicant and this reviewer were similar to those of the preceding UTI studies. Escherichia coli was the predominant pathogen in all three treatment groups. The applicant reported microbiologic cures of 83% (24/29) in the aztreonam group, and 100 % (13/13) in the gentamicin group, at 5-9 days after completion of therapy. Clinical cure was reported in 100% of both treatment groups. This reviewer's evaluation of the domestic and foreign data on complicated and uncomplicated UTI are presented in Table II (A-B). Bacteriologic responses for complicated and uncomplicated UTI in the domestic and foreign study populations are presented in Table II (A-C; E-G). The overall bacteriologic cure rates for complicated and uncomplicated UTI combined were 76.2% (32/42) in the aztreonam group and 73.3% (11/15) in the gentamicin group (Table II: I). All three pataients treated with netilmicin were bacteriologically cured. The bacteriologic cure rate was higher (86.7%) in the foreign study population, as compared to that (70.4%) in the domestic study population. The number of patients in each study, however, was small. The bacteriologic cure rates for the aztreonam-treatment group were similar to those in the preceding UTI studies. The reinfection rate was higher in the gentamicin group (46.7%) as compared to the aztreonam group (15.6%). The superinfection rates, however, were similar in the two treatment groups, 8.9% (4/30) in the aztreonam group and 8.3% (1/15) in the gentamicin group.

Superinfections were caused by S. faecalis in the aztreonam group, and by S. epidermidis in the gentamicin group. Two patients of the aztreonam group were treated with other antibiotics for superinfection. The clinical cure rates were similar in the two treatment groups, as shown in Table II (I).

The Safety of the test and control drugs (aminoglycosides) was evaluated in all 126 patients who were treated. Adverse reactions possibly or probably related to drug therapy were reported in 11 patients (12.8%) in the aztreonam group and in 3 patients(7.5%) in the aminoglycosides group. Drug therapy was discontinued in 4 patients, two in each treatment group, because of phlebitis in the aztreonam group, and renal dysfunction in the gentamicin group. A transient increase in transaminase (ALT/AST) levels was observed in 5.6% of the aztreonam group, whereas an increase in serum creatinine levels was observed in 8.6% of the aminoglycoside group. There were 7 deaths among the study population, 2 in the aztreonam group and 5 in the gentamicin group. The deaths were not attributed to drug therapy. The adverse reactions reported were as follows:

Number of Patients Treated	Aztreonam	Aminoglycosides
	86	40
Adverse Reactions		
Clinical:		
Nausea	6	2
Taste alteration	1	0
Renal failure/azotemia	1	0
Phlebitis/local reaction	0	2
Vaginitis (Candida)	4	0
	1	0
Laboratory:		
PT/PTT	6	3
ALT(SGPT)/AST(SGOT)	3 (61)	0
Creatinine	4 (71)	0
	0	3 (35)

Note: A few patients had more than one adverse reaction.
The numbers in parentheses represent the number of patients in whom the laboratory tests were done.

Conclusions: This multicenter, randomized, controlled study of aztreonam vs. aminoglycoside (gentamicin) indicated that the efficacy and safety of the test and the control drug were comparable in the treatment of hospitalized patients with gram-negative urinary tract infections. The bacteriologic cure rate of azteonam seen in this study was similar to that in the preceding UTI studies.

Table I (A)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 6218; 6250; 6337*; 7506

	Aztreonam	Gentamicin
Total Number of Patients Entered:	47	20
No. of Patients Not Evaluable for Efficacy	16	8
Reasons:		
Improper or negative pretreatment culture	4	1
Resistant organisms/Susceptibility unknown	2	0
Inappropriate follow-up	2	6
5 days of therapy	3	0
Surgical procedure or concurrent antimicrobial therapy	4	1
Clinical diagnosis other than UTI	1	0
Other (no evaluable patient in control group)	1	0
No. of Patients Evaluable for Efficacy*	30	12
Demographic Characteristics:		
Sex		
Female	18	7
Male	12	5
Age (Years)		
Range	16 - 80	23 - 75
Mean	53.7	57.5
Race		
Caucasian	27	11
Black	3	1
Clinical Diagnosis		
UTI (unspecified)	1	-
Pyelonephritis	7	2
Cystitis	22	10
Complicated UTI	19	6
Uncomplicated UTI	11	6
Duration of Treatment (days)		
Range	5 - 16	5 - 10
Mean	8.7	7.6

*The investigator did not enter evaluable patients into both treatment groups.
**Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table I (B)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Urinary Tract Infection

Foreign Study

Investigators' Number: 6187, 6240, 6308*, 6415*, 6419, 6436, 6459*

	Azt.	Gen	Net.
Total Number of Patients Entered:	39	15	5
No. of Patients Not Evaluable for Efficacy	24	12	1
Reasons:			
Improper or negative pretreatment culture	13	4	0
Inappropriate follow-up	2	1	0
5 days of therapy	0	1	1
Concurrent antimicrobial therapy	0	1	0
Other (no evaluable patient in control group)	3	0	0
Randomization not done	6	5	0
No. of Patients Evaluable for Efficacy*	15	3	4
Demographic Characteristics:			
Sex			
Female	10	2	3
Male	5	1	1
Age (Years)			
Range	32-72	42-74	39-76
Mean	57	56	56
Race			
Caucasian	15	3	4
Clinical Diagnosis			
Pyelonephritis	5	0	0
Cystitis/lower UTI	10	3	4
Complicated UTI	8	2	1
Uncomplicated UTI	7	1	3
Duration of Treatment (days)			
Range	5-11	8-10	7-10
Mean	7.5	9	8

*The investigator did not enter evaluable patients into both treatment groups.
**Patients who had appropriate follow-up: up to 4-6 weeks after completion of therapy.

Table II (A)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Complicated Urinary Tract Infection
Bacteriologic Response*

	Aztreonam					Gentamicin				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	3/7	1	3	0	1(a)	3/5	0	2	2(L)	0
K. pneumoniae	2/4	1	0	1(E)**	1**	0/0	-	-	-	-
K. oxytoca	1/1	0	0	0	0	0/0	-	-	-	-
E. cloacae	2/2**	0	0	2(L)**	2	0/0	-	-	-	-
C. diversus	1/1	0	0	0	0	0/0	-	-	-	-
P. aeruginosa	1/3	1	0	1(E)	0	1/1	0	0	0	0
P. aeruginosa + P. rettgeri	1/1	0	0	0	0	0/0	-	-	-	-
Total	11/19 (58%)	3	3	4(2-E) (2-L)	4	4/6	0	2	2(L)	0

* At 4-6 weeks after completion of therapy.
** The same patient
E - Eradicated: number eradicated/number treated
P - Persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection: (a) The original pathogen was eradicated.

Table II (B)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Uncomplicated Urinary Tract Infection
Bacteriologic Response*

	Aztreonam					Gentamicin				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	3/6	1	1	1(E)	0	1/3	2	0	2(1-L)	0
P. mirabilis	1/1	0	0	0	0	0/0	-	-	-	-
K. pneumoniae	0/0	-	-	-	-	2/2	0	0	1(L)**	1**
E. cloacae	1/1	0	0	0	0	0/0	-	-	-	-
P. aeruginosa	2/2	0	0	0	0	1/1	0	0	0	0
S. marcescens	1/1	0	0	0	0	0/0	-	-	-	-
Total	8/11 (73%)	1	1	1(E)	0	4/6	2	0	3(2-L) (1-E)	1

* At 4-6 weeks after completion of therapy.
** The same patient
E - Eradicated: number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy
SI - superinfection

Table II (C)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Urinary Tract Infection(Complicated + Uncomplicated)
Bacteriologic Response*

	Aztreonam					Gentamicin				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI
Single pathogen:										
E. coli	6/13	2	4	1(E)	1(a)	4/8	2	2	4(3-L)	0
P. aeruginosa	3/5	1	0	1(E)	0	2/2	0	0	0	0
K. pneumoniae	2/4	1	0	1(E)**	1**	2/2	0	0	1(L)**	1**
E. cloacae	3/3	0	0	2(L)(a)	2	0/0	-	-	-	-
C. diversus	1/1	0	0	0	0	0/0	-	-	-	-
K. oxytoca	1/1	0	0	0	0	0/0	-	-	-	-
P. mirabilis	1/1	0	0	0	0	0/0	-	-	-	-
S. marcescens	1/1	0	0	0	0	0/0	-	-	-	-
Multiple pathogens:										
P. aeruginosa + F. rettgeri	1/1	0	0	0	0	0/0	-	-	-	-
Total	19/30 (63%)	4	4	5(3-E) (2-L)	4	8/12 (67%)	2	2	5(4-L) (1-L)	1

* At 4-6 weeks after completion of therapy.
** The same patient
E - Eradicated: number eradicated/number treated
P - Persisted
RL - relapsed
RI - reinfected: (E) - reinfected within 5-9 days post-therapy
(L) - reinfected at 4-6 weeks post-therapy

SI - superinfection: (a) The original pathogen was eradicated.

Table II (D)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Urinary Tract Infection (Complicated + Uncomplicated)
Bacteriologic Cure (eradication of original pathogen)*

UTI	Number Cured/Number Treated** (cure rate)	
	Aztreonam	Gentamicin
Complicated	11/17	4/6
Uncomplicated	8/10	4/6
Total	19/27 (70.4%)	8/12 (66.7%)

Clinical Response (Cure + Improvement)*

UTI	Number Cured + Improved/Number Treated**	
	Aztreonam	Gentamicin
Complicated	14/17	5/5
Uncomplicated	10/10	5/5
Total	24/27 (88.9%)	10/10 (100%)

*At 4 - 6 weeks after completion of therapy
**Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Table II (E)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Complicated Urinary Tract Infection
Bacteriologic Response*

	Aztreonam					Gentamicin					Netilmicin				
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	3/3	0	0	0	0	1/1**	0	0	1(L)**	0	0/1	0	0	1(E)	0
P. mirabilis	2/2	0	0	0	0	0/0	-	-	-	-	0/0	-	-	-	-
K. pneumoniae/ Klebsiella sp.	1/1	0	0	0	0	1/1	0	0	0	0	0/0	-	-	-	-
P. aeruginosa	0/1	1	0	0	0	0/0	-	-	-	-	0/0	-	-	-	-
Serratia sp.	1/1**	0	0	1(L)**	0	0/0	-	-	-	-	0/0	-	-	-	-
Total	7/8	1	0	1(L)	0	2/2	0	0	1(L)	0	0/1	0	0	1	0

* At 4-6 weeks after completion of therapy.
** The same patient
E - Eradicated: number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected
SI - superinfection
(E) - reinfection at 5-9 days post-therapy
(L) - reinfection at 4-6 weeks post-therapy

Table II (F)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Uncomplicated urinary Tract Infection
Bacteriologic Response*

Pathogen	Aztreonam					Gentamicin					Netilmicin				
	E	P	RL	RI	SI	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	4/4	0	0	1(L)	0	0/0	-	-	-	-	2/2	0	0	1(E)	0
Proteus sp.	1/1	0	0	0	0	0/0	-	-	-	-	0/0	-	-	-	-
K. pneumoniae	1/1	0	0	0	0	0/0	-	-	-	-	0/0	-	-	-	-
K. oxytoca	0/0	-	-	-	-	0/0	-	-	-	-	1/1	0	0	0	0
E. cloacae	0/1	0	1	0	0	0/0	-	-	-	-	0/0	-	-	-	-
C. freundii	0/0	-	-	-	-	1/1**	0	0	1(L)**	-	0/0	-	-	-	-
Total	6/7	0	1	1(L)	0	1/1	0	0	1(L)	0	3/3	0	0	0	0

* At 4-6 weeks after completion of therapy.
** The same patient
E - Eradated: number eradicated/number treated
P - persisted
RL - relapsed
RI - reinfected
SI - superinfection
(E) - reinfection at 5-9 days post-therapy
(L) - reinfection at 4-6 weeks post-therapy

Table II (G)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

	Urinary Tract Infection (Complicated + Uncomplicated)															
	Bacteriologic Response*															
	Aztreonam					Gentamicin					Netilmicin					
Pathogen	E	P	RL	RI	SI	E	P	RL	RI	SI	E	P	RL	RI	SI
E. coli	7/7	0	0	1(L)	0	1/1**	-	-	1(L)**	0	2/3	0	0	1(E)	0
Proteus sp./ P. mirabilis	3/3	0	0	0	0	0/0	-	-	-	-	0/0	-	-	-	-
K. pneumoniae/ Klebsiella sp.	2/2	0	0	0	0	1/1	-	-	-	-	0/0	-	-	-	-
K. oxytoca	0/0	-	-	-	-	0/0	-	-	-	-	1/1	0	0	0	0
E cloacae	0/1	0	1	0	0	0/0	-	-	-	-	0/0	-	-	-	-
C. freundii	0/0	-	-	-	-	1/1**	0	0	1(L)**	0	0/0	-	-	-	-
P. aeruginosa	0/1	1	0	0	0	0/0	-	-	-	-	0/0	-	-	-	-
Serratia sp.	1/1**	0	0	1(L)**	0	0/0	-	-	-	-	0/0	-	-	-	-
Total	13/15 (87%)	1	1	2(L)	0	3/3	0	0	2(L)	0	3/4	0	0	1	0

* At 4-6 weeks after completion of therapy.
** The same patient
E - Eradicated: number eradicated/number treated
P - Persisted
RL - relapsed
RI - reinfected
SI - superinfection
(E) - reinfection at 5-9 days post-therapy
(L) - reinfection at 4-6 weeks post-therapy

Table II (H)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Foreign Study

Urinary Tract Infection (Complicated + Uncomplicated)
Bacteriologic Cure (eradication of original pathogen)*

UTI	Number Cured/Number Treated** (cure rate)		
	Aztreonam	Gentamicin	Netilmicin
Complicated	7/8	2/2	0/0
Uncomplicated	6/7	1/1	3/3
Total	13/15 (86.7%)	3/3	3/3

UTI	Clinical Response (Cure + Improvement)* Number Cured + Improved/Number Treated**		
	Aztreonam	Gentamicin	Netilmicin
Complicated	7/8	1/2	0/0
Uncomplicated	5/7	1/1	3/3
Total	12/15 (80%)	2/3	3/3

*At 4 - 6 weeks after completion of therapy
**Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Table II (I)

Protocol 18554-27: Comparison of Aztreonam vs. Aminoglycosides in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic and Foreign Studies Pooled:

Urinary Tract Infection (Complicated + Uncomplicated)
Bacteriologic Cure (eradication of original pathogen)*

UTI	Number Cured/Number Treated** (cure rate)		
	Aztreonam	Gentamicin	Netilmicin
Complicated	18/25 (72%)	6/8	0/0
Uncomplicated	14/17 (82%	5/7	3/3
Total	32/42 (76.2%)	11/15 (73.3%)	3/3

Clinical Response (Cure + Improvement)*

UTI	Number Cured + Improved/Number Treated**		
	Aztreonam	Gentamicin	Netilmicin
Complicated	21/25 (84%)	6/7	0/0
Uncomplicated	15/17 (88%)	6/6	3/3
Total	36/42 (85.7%)	12/13 (92.3%)	3/3

*At 4 - 6 weeks after completion of therapy
**Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Protocol 18554-28: Comparison of Aztreonam vs. Tobramycin in the Treatment of Serious Gram-negative Urinary Tract Infections

This was a single investigator, domestic study of aztreonam vs. an aminoglycoside, tobramycin. in the treatment of hospitalized patients with gram-negative UTI.

Patients with a presumptive diagnosis of UTI were entered into this study, as in the preceding UTI studies. Appropriate urine cultures were done to confirm the diagnosis, and susceptibility testing of clinical isolates was performed by the disc method. The procedures for the monitoring of patients during and after completion of therapy were similar to those in the preceding multicenter UTI studies.
The dosage regimen of aztreonam was the same as that used in the preceding UTI studies, except that the drug was administered intramuscularly in all patients. The dosage of tobramycin was 1 mg/kg every 8 hours. The duration of therapy ranged from 5 to 10 days.

Of a total of 49 patients entered into this study, 33 were allocated to the aztreonam group and 16 to the tobramycin group, using a randomization ratio of 2:1, respectively. All patients were males. The demographics of the two treatment groups were similar with respect to age, weight, and status of UTI (complicated). Twenty patients, 11 from the aztreonam group and 9 from the tobramycin group, were excluded from the efficacy evaluation. The reasons for exlcusion are presented in Table I (A). The demography of the evaluable patient remained similar in the two treatment groups. Urologic surgery was performed during therapy in 6 patients of the aztreonam group and in 3 patients of the tobramycin group. In this study, Pseudomonas aeruginosa was the predominant pathogen, followed by E. coli.

The applicant reported bacteriologic cure rates of 71% (20/28)and 88% (7/8) for the aztreonam group and the tobramycin group, respectively at 5-9 days post-therapy. This reviewer's evaluation at 4-6 weeks post-therapy showed bacteriologic cure rates of 55% (11/20) and 66.7% (4/6), respectively. The lower cure rates for both treatment groups were not unexpected since the cure rates for complicated UTI in males are usually lower with currently marketed antimicrobials, especially for UTI caused by P. aeruginosa. Superinfection occurred in 2 patients (9%) in the aztreonam group, and in none in the tobramycin group. One of the two patients were treated with an antibiotic, ampicillin. Streptococcus faecalis was the organism causing superinfection in both patients. Reinfection occurred in 1 patient (4.5%) of the aztreonam group and in 2 (25%) of the tobramycin group. The causative organisms were Enterobacter aerogenes and Streptococcus epidermidis, respectively, and the microoganisms were susceptible to the treatment drugs. The number of patients in this study was small; nevertheless, the results of this study were similar to those found in the preceding multicenter UTI studies.

The safety of the test and control drugs were evaluated in all patients who received the drugs, 33 in the aztreonam group and 16 in the tobramycin group. Adverse reactions, possibly or probably related to drug therapy were reported in 12 aztreonam-treated patients (36%) and in 4 tobramycin-treated patients (25%). In none of the patients was discontinuance of therapy necessary. Laboratory abnormalities, primarily transient increases in transaminase (ALT/AST) levels were observed in 7 (21%) of the aztreonam group and 3 (15.8%) of the tobramycin group. All of the tranaminase levels were below 100 IU/ml. The advers reactions observed were as follows:

Number of Patients Treated Adverse Reactions	Aztreonam 33	Aminoglycosides 16
Clinical:	2	0
Diarrhea	1	0
Pain at injection site	1	0
Laboratory abnormalities:	10	4
Eosinophilia	2 (26)	1 (13)
ALT(SGPT)/AST(SGOT)	7 (33)	3 (16)
Serum potassium*	1 (30)	0 (14)

The numbers in parentheses represent the number of patients in whom the laboratory tests were done.

Conclusions: This randomized, controlled study of aztreonam vs. tobramycin, an aminoglycoside, by a single domestic investigator indicated that aztreonam is as effective and safe as the control drug in the treatment of complicated UTI caused by gram-negative uropathogens in this small study population.

Form 5 50-580

Table I (A)

Protocol 18554-28: Comparison of Aztreonam vs. Tobramycinin the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 2890

	Aztreonam	Tobramycin
Total Number of Patients Entered:	33	16
No. of Patients Not Evaluable for Efficacy	11	9
Reasons:		
Improper or negative pretreatment culture	2	3
Inappropriate follow-up	4	1
5 days of therapy	1	0
Surgical procedure during follow-up or concurrent antimicrobial therapy	1	3
Clinical diagnosis other than UTI	3	1
Other (no evaluable patient in control group)	1	0
No. of Patients Evaluable for Efficacy*	22	7
Demographic Characteristics:		
Sex		
Male	22	7
Age (Years)		
Range	52 - 89	56 - 78
Mean	69.7	65
Race		
Caucasian	22	8
Clinical Diagnosis		
Pyelonephritis	3	0
Cystitis	19	8
Complicated UTI	22	8
Duration of Symptoms (days):		
Range	1 - 15	1 - 4
Mean	4.1	2.3
Dosage Regimen:(IM; q 8 h)	0.5 - 1 g	76 -80 mg
Duration of Treatment (days)		
Range	5 - 10	7 - 8
Mean	7.3	7.3

*The investigator did not enter evaluable patients into both treatment groups.
**Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table II (B)

Protocol 18554-28: Comparison of Aztreonam vs. Tobramycin in the Treatment of Serious Gram-negative Urinary Tract Infection

Domestic Study

Investigators' Number: 2890

Complicated Urinary Tract Infection
Bacteriologic Response *

Pathogen	Aztreonam E	P	RL	RI	SI	Tobramycin E	P	RL	RI	SI
Single pathogen:										
P. aeruginosa	1/7	3	2	1(E)**	1**	1/2	0	0	2(1-E)	0
E. coli	3/5	2	0	0	0	1/2	0	1	0	0
C. freundii/ Citrobacter sp.	1/2	0	0	0	1	0/0	-	-	-	-
E. cloacae	1/1	0	0	0	0	1/1	0	0	0	0
K. oxytoca	1/1	0	0	0	0	1/1	0	0	0	0
E. aerogenes	0/1	1	0	0	0	0/0	-	-	-	-
M. morganii	1/1	0	0	0	0	0/0	-	-	-	-
P. stuartii	0/1	1	0	0	0	0/0	-	-	-	-
S. marcescens	1/1	0	0	0	0	0/0	-	-	-	-
Multiple pathogens:										
K. pneumoniae + E. cloacae	1/1	0	0	0	0	0/0	-	-	-	-
P. mirabilis + M. morganii	0/0	-	-	-	-	0/1	1	0	0	0
Total	11/22 (50%)	7	2	1(E) (1-L)	2	4/7	1	1	2(1-E) (1-L)	0

* At 4-6 weeks after completion of therapy ** The same patients
E- eradication, number eradicated/number treated
P - persistence
RL - relapse
RI - reinfection
SI - superinfection
(E) - Reinfection at 5-9 days after completion of therapy
(L) - Reinfection at 4-6 weeks after completion of therapy

Protocol 18-554-11: Comparison of Aztreonam and Tobramycin in the Treatment of Lower Respiratory Tract Infection (LRTI) due to Aerobic Gram-negative Microorganisms

This was a multicenter, randomized study of aztreonam vs tobramycin, an aminoglycoside, in the treatment of hospitalized patients with aerobic gram-negative LRTI. The exclusion criteria were similar to those in other controlled clinical studies of aztreonam. (see Appendix)

Sixteen principal investigators, 11 domestic and 5 foreign, entered a total of 247 patients (173 domestic and 74 foreign) into this study. A presumptive diagnosis of LRTI was made in the presence of clinical symptoms and signs, and roentgenologic findings compatible with pneumonia or bronchitis. Microbiologic examination of purulent sputum or tracheal aspirate was done for quantitative analyses of polymorphonuclear leukocytes (PMN) and squamous epithelial cells per low-power field. A pretherapy sputum containing $\geq$25 PMN leukocytes and $<$ 10 squamous epithelial cells/LPF was considered to be indicative of LRTI. The diagnosis was confirmed by appropriate sputum cultures. Susceptibility testing of clinical isolates was performed, using the disc method or tube dilution method. Routine physical examination, laboratory tests (blood counts, blood chemistries, and urinalyses), and chest roentgenogram were done prior to, during, and/or after completion of therapy. The patients were randomly allocated in a ratio of 2:1 to receive either aztreonam or tobramycin, respectively. Patients assigned to tobramycin therapy from whom Haemophilus influenzae was isolated were treated with moxalactam, a third-generation cephalosporin, since tobramycin has little activity against this pathogen. The investigators were not blinded. Dosage regimens were 1 or 2 g of aztreonam q 8 h , 80 mg of tobramycin q 8 h, and 1 or 2 g of moxalactam q 8 h. In the majority of patients the drugs were administered intravenously. A few patients received aztreonam and tobramycin intramuscularly. Concomitant use of clindamycin or other antibiotics with activity against anaerobic and/or gram-positive organisms was allowed in this study. The duration of therapy ranged from 5 to 15 days in the majority of patients.

Of the 247 patients entered into this study, 173 patients (128 domestic and 45 foreign) received aztreonam, 68 patients (55 domestic and 13 foreign) received tobramycin, 3 patients (domestic) received moxalactam, 1 domestic patient received both tobramycin and moxalactam, and the remaining 2 patients (foreign) received no drug. The demographic characteristics of the three treatment groups were presented by the applicant in its Tables 2 and 3 (vol. 3.3: 2874-5). The demography of all patients treated with aztreonam and tobramycin was similar, with respect to age, weight, sex, and race. Seventy-six percent of the aztreonam-treated patients, 78% of tobramycin-treated patients, and two of the four moxalactam-treated patients were male. The mean age of patients was 65 in the aztreonam group and 66 in the tobramycin group, and the mean weights were 65 kg and 66 kg, respectively. Ninety-five percent in the aztreonam group, and 93% in the tobramycin group were Caucasian.

One-hundred-twenty one patients (78 domestic and 43 foreign) were excluded from the efficacy evaluation for reasons listed in this reviewer's Table I (A)-domestic studies- and Table II (A)- foreign studies. The major reasons for exclusion were a failure to isolate aerobic gram-negative microorganisms from appropriate specimens. The demography of domestic and foreign patients who were evaluable for efficacy is presented in Table I (A) and Table II (A). The demographics of the evaluable patients in the two treatment groups remained similar to that of all treated patients presented by the applicant.

This reviewer excluded data from 7 investigators (4 domestic and 3 foreign) who did not have evaluable patients in both treatment groups. The number of evaluable patients was therefore smaller than that presented by the applicant. The primary clinical diagnosis of the evaluable patients was pneumonia in 94 % of the aztreonam-treated patients and 91% of the tobramycin-treated patients. One patient in both treatment groups had a lung abscess. The distribution of patients with multiple predisposing conditions (chronic obstructive pulmonary disease, congestive heart failure or pulmonary carcinoma) was similar in the two treatment groups.

As in the evaluation of the preceding clinical studies, the applicant pooled the results of domestic and foreign studies in its analyses of efficacy data. In the evaluation of microbiologic responses, the applicant stated that 33% of patients had sputum indicative of LRTI. It reported that the microbiologic and clinical cure rates for each drug were similar in patients whose sputum cell counts met the criteria, those in whom it did not, and those for whom sputum cell counts were not reported; the rates for microbiologic eradication were 90%, 84%, and 82%, respectively, in the aztreonam group, and 73%, 69% and 75%, respectively in the tobramycin group. The distribution of organisms and cell count categories were analysed by the applicant, as shown in its Table 11 (vol. 3.3: 2884-5). The distribution of pathogens was similar among the treatment groups. The applicant therefore analysed results of all evaluable patients, regardless of whether the sputum samples met the cell count criteria, as originally defined in its protocol. The review of the patient case reports revealed that two-thirds of the evaluable patients in the test and control groups had sputum findings indicative of LRTI. As stated in the applicant's summary tables for the efficacy evaluation in LRTI, an assumption of microbiologic eradication was made when clinical improvement was shown by resolution of symptoms and signs of LRTI, including absence of sputum production during and/or after completion of therapy. The protocol for this LRTI study called for post-therapy follow-ups at 1-5 days and 3-4 weeks after completion of therapy. Only one-third of the evaluable patient had the 2-4 week post-therapy follow-up.

Two-thirds of the evaluable patients in both (aztreonam and tobramycin) groups in the domestic studies received clindamycin concurrently, whereas one patient in the aztreonam group in the foreign studies received clindamycin

As shown in Tables I(A) and II(A), 86% (66/77) of the aztreonam group and 69% (20/29) in the tobramycin group had LRTI caused by single aerobic gram-negative pathogens. Multiple gram-negative pathogens (2 or more) were causative in the remaining patients. The microbiologic responses seen in the test and control groups are shown in Tables I (A) and II (A). Pseumononas aeruginosa, Escherichia coli, Klebsiella pneumoniae, Haemophilus influenzae, and Enterobacter species, including E. cloacae and E. aerogenes were the predominant pathogens in the study population.

In the evaluation of microbiologic responses, this reviewer considered the pathogens not eradicated when the initial pathogens were isolated again at post-therapy follow-up (relapse). As in the evaluation of UTI studies, such cases were considered as a microbiologic failure rather than a 'microbiologic cure with relapse. Although microbiologic and clinical cures were well defined in the protocol of this study, the evaluation of the data were compromised by the fact that only one-third of the patients had adequate post-therapy follow-up. This was particularly pertinent in the evaluation of the patients with underlying chronic obstructive pulmonary diseases (COPD). An appropriate interpretation of the response appeared to be clinical improvement, rather than clinical cure. For the same reason, interpretation of the microbiologic results (eradication, relapse, and reinfection) was limited. The results of this multicenter study, nevertheless, indicated that aztreonam was as effective as the control drug, tobramycin, in the treatment of LRTI caused by aerobic gram-negative pathogens, when an effective antianaerobic drug was used concomitantly. The overall microbiologic eradication rate in the domestic studies were 76.6% (59/77) in the aztreonam group and 62.1% (18/29) in the tobramycin group. In the foreign studies, the eradication rate was 83.3 % (10/11) in the aztreonam group. Two of the three patients in the foreign studies had a favorable microbiologic response, and 3 of the 4 patients treated with moxalactam also had a favorable response. Overall clinical improvement was seen in 87% (67/77) of the aztreonam group and in 69% (20/29) of the tobramycin group in the domestic studies. As expected, patients with single gram-negative pathogens appeared to have a better microbiologic and clinical outcome than those with multiple gram-negative pathogens. Pseudomonas aeruginosa was the most difficult pathogen to eradicate in the test and the control groups, whcih was not unexpected. Superinfections occurred in 15 patients (19.5%) in the aztreonam group and in 8 patients (27.6%) in the tobramycin group in the domestic studies. The microorganisms were Streptococcus pneumoniae in 4, Pseudomonas maltophila in 3, S aureus + Klebsiella oxytoca in 1, and one each of E. cloacae, Staphylococcus aureus, S. epidermidis, P. fluorescens, and Acinetobacter calcoaceticus in the aztreonam group, and one each of P. aeruginosa + E. aerogenes, E. cloacae + K. oxytoca, E. coli + P. mirabilis + P. maltophilia, P. aeruginosa + E. coli, S. marcescens, E. coli + monilia in the tobramycin group. Nine of the 15 aztreonam-treated patients and 4 of the 8 patients in the tobramycin group were treated with other antibiotics for superinfection.

Safety was evaluated in all patients who received the test and control drugs. Adverse effects (clinical and laboratory), possibly or probably related to drug therapy were observed in 25 (20%) of the 128 aztreonam-treated patients and in 11 (20%) of the 55 tobramycin-treated patients in domestic studies, and in 1 (22%) of the 45 aztreonam-treated patients and 2 (15.4%) of the 13 tobramycin-treated patients in foreign studies. Drug therapy was discontinued in 3 aztreonam-treated patients, and in 2 tobramycin-treated patients because of adverse reactions. The incidences of clinical and laboratory adverse effects were similar in the two treatment groups (aztreonam and tobramycin).

The adverse reactions observed were as follows:

	Aztreonam		Tobramycin		Moxalactam	
Total Number of Patients Treated	128(D)	45(F)	55(D)	13(F)	4*(D)	0(F)
Adverse Reactions						
Clinical:	11	1	3	0	0	0
Nausea	4	0	0	0	0	0
Diarrhea	1	0	1	0	0	0
Rash	3	0	2	0	0	0
Purpura	0	1	0	0	0	0
Phlebitis	2	0	0	0	0	0
Laboratory abnormalities:	16 (D+F)		9 (D+F)		1 (D+F)	
Eosinophilia	5 (143)		1 (62)		0	
Thrombocytopenia	1 (123)		0 (57)		0	
Thrombocytosis	1 (123)		0 (57)		0	
PT/PTT	0 (126)		0 (54)		1 (4)	
ALT(SGPT)/AST(SGOT)	6 (125)		3 (63)		0	
Alkaline phosphatase	2 (148)		1 (63)		0	
BUN/creatinine	1 (148)		4 (63)		0	

The numbers in parentheses represent the number of patients in whom the laboratory tests were done.
D- Domestic studies
F- Foreign studies.

Deaths: In this severely ill study population, there were 46 deaths: 35 (20%) in the aztreonam group, 9 (13%) in the tobramycin group, and 2 in the moxalactam group. One of the two patients in the latter group also received tobramycin. The deaths were not attributed to the drug therapy. A summary of the cases with fatal outcome was presented by the applicant in volume 3.4 (pp 4-11).

Conclusions: This multicenter, randomized, controlled study of aztreonam vs. an aminoglycoside tobramycin indicated that the efficacy and safety of the test and the control drug were comparable in the treatment of hospitalized patients with aerobic gram-negative lower respiratory tract infections(LRTI).

TABLE I

Protocol No. 18554-11: Comparison of Aztreonam and Tobramycin in the Treatment of Aerobic Gram-negative Lower Respiratory Tract Infections.

Domestic Study

Number of Investigators: 11*

Investigator's Numbers: 3096, 6207, 6226, 6227, 6228, 6317*, 6449, 7614, 6224*, 6229*, 6401*

	Aztreonam	Tobramycin	Moxalactam
Total Number of Patients Entered:	128	55	4
No. of Patients Not Evaluable for Efficacy	51	26	0
Reasons: Bacteriological criteria not met	39	23	-
> days of therapy	5	-	-
Concurrent antimicrobial therapy	1	2	-
No evaluable patients in the control group	6	-	-
Incomplete data	-	1	-
No. of Patients Evaluable for Efficacy*	77	29	4
Demographic Characteristics:			
Sex			
Female	21	7	2
Male	56	22	2
Age			
Range	32 - 93	22 - 92	54 - 84
Mean	65	69	68
Race			
Caucasian	72	27	4
Black	4	2	0
Other	1	0	0
Clinical Diagnosis			
Pneumonia	73	28	3
Bronchitis	4	1	1
(Concurrent Cardiopulmonary Disease	16	6	1)
Dosage Regimens (IV or IM q 8 h)	1 - 2g	60 - 90 mg	2g
Duration of Treatment (Days)			
Range	5 - 27	5 - 15	5 - 11
Mean	10	9.3	8.3
Concomitant Antibiotics Used (Clindamycin)	57	21	1

*4 investigators had no evaluable patients in the control group.

TABLE I(A)

Protocol No. 18554-11: Comparison of Aztreonam and Tobramycin in the Treatment of Aerobic Gram-negative Lower Respiratory Tract Infections.

Domestic Study:

Microbiologic Response

Pathogen	No. Eradicated*/No. of Patients Treated		
	Aztreonam	Tobramycin	Moxalactam
Single Pathogen:			
P. aeruginosa/ Pseudomonas sp.	12/19	3/8	-
E. coli	13/13	1/1	-
K. pneumoniae	8/9	4/4	-
H. influenzae	7/8	1/1	1/1
E. aerogenes	3/4	-	-
Enterobacter sp.	2/2	1/1	-
E. cloacae	1/1	2/2	-
K. oxytoca	1/2	-	-
Serratia sp.	2/2	1/1	-
P. mirabilis	2/2	0/1	-
P. stuartii	1/1	-	-
M. morganii	1/1	-	-
S. rubidaea	1/1	-	-
C. diversus	1/1	1/1	-
H. parainfluenzae	1/1	-	1/1**
Total	55/66 (83.3%)	14/20 (70.0%)	2/2

*Microbiological eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.
**This patient was treated with tobramycin and moxalactam.

TABLE I(A) continued

Pathogen	No. Eradicated*/No. of Patients Treated		
	Aztreonam	Tobramycin	Moxalactam
Multiple Pathogens:			
E. coli + Acinetobacter sp.	-	1/1	-
E. coli + E. aerogenes	1/1	1/1	-
E. coli + P. mirabilis***	0/1	-	-
H. influenzae + K. pneumoniae	-	-	1/1
H. influenzae + K. oxytoca	-	-	1/1
H. influenzae + P. mirabilis***	-	0/1	-
K. pneumoniae + E. cloacae	1/1	-	-
K. pneumoniae + P. mirabilis	-	1/1	-
K. pneumoniae + P. aeruginosa****	1/4	-	-
P. mirabilis + E. cloacae	1/1	-	-
P. mirabilis + P. aeruginosa***	-	0/1	-
P. aeruginosa*** + K. oxytoca	0/1	-	-
P. aeruginosa + E. cloacae	0/1	-	-
K. oxytoca + E. cloacae	1/1	-	-
K. pneumonia*** + E. aeruginosa + E. cloacae	-	0/1	-
K. pneumoniae + P. mirabilis + K. oxytoca	-	1/1	-
P. mirabilis*** + E. coli *** + E. cloacae	-	0/1	-
P. mirabilis + S. marcescens + P. aeruginosa	-	0/1	-
Total	4/11	4/9	2/2
Total(single + multiple)	59/77 (76.6%)	18/29 (62.1%)	4/4

*Microbiological eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.
***The organisms not eradicated.
**** Pseudomonas aeruginosa was the pathogen not eradicated in 3 of the 4 patients.

TABLE II

Protocol No. 18554-11: Comparison of Aztreonam and Tobramycin in the Treatment of Aerobic Gram-negative Lower Respiratory Tract Infections.

Foreign Study

Number of Investigators: 5*

Investigator's Numbers: 6345, 6357*, 6358*, 6366, 6429*

	Aztreonam	Tobramycin
Total Number of Patients Entered:	45	13
No. of Patients Not Evaluable for Efficacy	34	10
Reasons: Bacteriological criteria not met	14	5
Inappropriate follow-up culture	3	0
Concurrent antimicrobial therapy	1	0
No evaluable pts. in the control group	2	0
Randomization not done	14	5
No. of Patients Evaluable for Efficacy*	11	3
Demographic Characteristics:		
Sex		
Female	1	1
Male	10	2
Age		
Range	16 - 79	47 - 80
Mean	65	69
Race		
Caucasian	11	3
Black	-	-
Other	-	-
Clinical Diagnosis		
Pneumonia	6	1
Bronchitis	4	1
Lung abscess	1	1
(Underlying pulmonary disease-COPD)	2	1
Dosage Regimens (IV)	0.5 -2 q 8 h	80 - 100 mg q 8 h
Duration of Treatment (Days)		
Range	5 - 21	9 - 11
Mean	11	10
Concomitant Antibiotics Used (Clindamycin)	1	0

*3 investigators had no evaluable patients in the control group.

TABLE II(A)

Foreign Study

Microbiologic Response

Pathogen	No. Eradicated*/No. of Patients Treated	
	Aztreonam	Tobramycin
Single Pathogen:		
E. coli	2/3	-
P. aeruginosa	2/2	1/2
H. influenzae	1/1	-
K. pneumoniae	1/1	-
Multiple Pathogens:		
H. Influenzae + K. pneumoniae	1/1	-
P. mirabilis + H. influenzae	-	1/1
P. mirabilis + S. marcescens	1/1	-
P. aeruginosa + H. influenzae	1/1	-
H. influenzae + P. vulgaris	1/1	-
Total	10/11 (83.3%)	2/3

*Microbiological eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Intra-abdominal Infections

This was a multicenter, randomized, comparative study of aztreonam vs. tobramycin in the treatment of intra-abdominal infections. Four domestic investigators and one foreign investigator participated in this study. A total of 60 patients were entered into the study by the domestic investigators, and six patients were entered by the foreign investigators, as presented in the following Tables. The patients were randomly assigned to either the aztreonam or the tobramycin group in a one to one ratio. The investigators were informed of the assigned drugs. Clindamycin was concurrently administered to all patients to cover anaerobes and gram-positive microorganisms. The treatment regimens were aztreonam 1-2 g q 8 - 12 h, or tobramycin, 3 mg/kg/day in two to three divided doses, and clindamycin 600 mg q 6 - 8 h. All of the study drugs were administered intravenously. The duration of therapy ranged from 6 to 21 days. The clinical and bacteriological diagnoses of intra-abdominal infections were made according to appropriate criteria set in the protocol. The majority of patients were diagnosed to have peritonitis. Susceptibility testing of aerobic gram-negative clinical isolates was done using the disc diffusion and/or tube dilution method. In a few patients the MICs and zone diameters were not recorded, but described only as 'sensitive' or 'resistant' by the investigators. A number of patients had polymicrobial infections, including obligate anaerobes which were expected in the intra-abdominal infections.

As presented in Table I(A) on the domestic studies, 33 of the 60 patients, 20 in the aztreonam group and 13 in the control group, were excluded from the efficacy evaluation by this reviewer. The reasons for exclusion are listed in the same table. The major reason was a negative culture for aerobic gram-negative microorganisms. The data from two domestic investigators who had less than one evaluable patient in each treatment group were excluded by this reviewer from the final analyses of efficacy. One investigator (6407) entered the great majority of the evaluable patients. The applicant, however, pooled domestic and foreign data, irrespective of the number of patients who were eligible for the efficacy evaluation in each treatment group. The total number of evaluable patients was quite small. Demography of the evaluable patients was comparable in the two treatment groups, as listed in Table I(A). The number of the patients who underwent surgical intervention prior to and during antimicrobial therapy was also comparable in the two treatment groups. The microbiological results are presented in Table I(B). Microbiological cure was seen in 10 (83.3%) of the 12 aztreonam-treated patients and in 8 (53.3%) of the 15 tobramycin-treated patients. Superinfection occurred in one (8.3%) of the twelve patients in the aztreonam group and one (6.7%) of the fifteen patients in the tobramycin group. The superinfection was due to S. epidermidis and S. faecalis in the aztreonam-treated patient, and S. epidermidis in the tobramycin-treated patient. The clinical cure rates were 83.3 % and 93.3%, respectively.

The safety of the drugs was assessed in 60 patients who were treated with the test or control drug.
Adverse reactions which were possibly or probably related to drug therapy were observed in 8 (25%) of the 32 patients in the aztreonam group and in 7 (25%) of the 28 patients in the tobramycin group. Two patients in each treatment group had more than one adverse reaction. In one patient of the aztreonam group, the drug was discontinued after 2 days of therapy. The reactions observed were as follows:

Number of Patients Treated	AZT/CLI 32	TOB/CLI 28
Clinical:		
Nausea and/or vomiting	3	4
Flatulence	0	1
Rash	2*	0
Laboratory abnormalities:		
Eosinophilia	1 (31)	2 (25)
Elevated AST(SGOT)/ALT(SGPT)	1 (25)	0 (22)
Prolonged PT/PTT	4** (29)	2** (16)

AZT - Aztreonam TOB - Tobramycin CLI - Clindamycin
* In one of the two patients the drug was discontinued.
** All patients received vitamin K.
The numbers in parentheses represent the number of patients in whom the laboratory tests were done.

Deaths occurred in 10 patients, 7 in the aztreonam group and 3 in the tobramycin group, during and after therapy. The deaths were not attributable to the drugs.

One foreign investigator (6444) from Brazil entered six patients into this multicenter study. As presented in Table II, all of the six patients, four in the aztreonam group and 2 in the tobramycin group, were evaluable for efficacy. The foreign study population was younger than the domestic study population. Only one patient in the test-drug group had surgical intervention during therapy. Microbiological and clinical cures were seen in all patients. No adverse reactions were reported in this foreign study.

Conclusions: Results of this multicenter, randomized study indicated that aztreonam, as an adjunct to surgery, appeared to be as effective and safe as the control drug, tobramycin, in the treatment of intra-abdominal infections caused by aerobic gram-negative pathogens, when these drugs were concomitantly used with an effective antianaerobic drug, clindamycin. The number of patients studied, however, was rather small, and therefore, more data are needed to confirm the results seen in this limited study.

Table I(A)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus Clindamycin in the Treatment of Intra-abdominal Infections (Domestic Study)

Number and (ID No.) of Principal Investigators: 4 (5099*;5766*,6407,6424)

	Treatment AZT + CLI	TOB + CLI
Total No. of Patients Entered:	32	28
Number of Patients Excluded from Efficacy Evaluation:	20	13
Reasons for Exclusion:		
Bacteriologic criteria not met (No gram-negative pathogens; inappropriate cultures)	18	13
Less than 5 days of therapy due to adverse reaction	1	0
Other (No evaluable patients in the control group)	1	0
Number of Patients Evaluable for Efficacy:	12	15
Demography and Other Characteristics of Evaluable Patients:		
Sex		
Female	7	8
Male	5	7
Age (years)		
Range	18 - 89	19 - 87
Mean	66.2	64.7
Race		
Caucasian	12	15
Clinical Diagnosis:		
Peritonitis	9	11
Abdominal inf. (ruptured appendix etc)	3	4
Surgery Prior to Antimicr al Therapy	3	6
Surgery During Antimicrobial Therapy	9	7
Dosage:		
Range	36 -140 g	0.96 - 4.8 g
Mean	68.8 g	2.9 g
Duration of Treatment (days):		
Range	6 - 21	8 - 20
Mean	12.1	12.3

*The investigator did not enter evaluable patients into both treatment groups.
AZT - aztreonam TOB - tobramycin CLI - clindamycin

Table I(B)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Intra-abdominal Infection (Domestic Study)

Microbiologic Response*

Gram-negative Pathogen	AZT + CLI	TOB + CLI
	No. Eradicated/No. Treated	
Single Pathogen:		
E. coli	5/5	4/7
P. mirabilis	1/1	1/2
E. cloacae	0/1	-
E. aerogenes	-	0/1
Multiple Pathogens:		
E. coli + K. pneumoniae	1/1	1/2
E. coli + P. vulgaris	1/1	-
E. coli + Ps. aeruginosa	-	1/1
K. pneumoniae + C. freundii	0/1	-
E. coli + K. pneumoniae + P. mirabilis	-	1/1
E. coli + K. pneumoniae + Ps. aeruginosa	1/1	0/1
E. coli + E. cloacae + Ps. aeruginosa + S. liquefaciens	1/1	-
Total	10/12 (83.3%)	7/13 (53.8)
Superinfection:		
S. epidermidis	0/12	1/15
S. epidermidis + S. faecalis	1/12	0/15

*Includes organisms of which the MICs and/or zone diameters were not recorded, but were reported as 'sensitive' to the test and control drugs by the investigators.

AZT - aztreonam
CLI - clindamycin
TOB - tobramycin

Table II(A)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Intra-abdominal Infection (Foreign Study)

No. of Principal Investigator and Investigator's Number: 1 , 6444

	Treatment AZT + CLI	TOB + CLI
Total No. of Patients Entered:	4	2
Number of Patients Excluded from Efficacy Evaluation:	0	0
Number of Patients Evaluable for Efficacy:	4	2
Demographic Characteristics of Evaluable Patients:		
Sex		
Female	4	1
Male	0	1
Age (years)		
Range	29 - 30	23 - 25
Mean	29.5	24.0
Race		
Caucasian	3	2
Black	1	0
Clinical Diagnosis		
Peritonitis	2	1
Abdominal inf. (ruptured appendix)	2	1
Surgery during Therapy	1	0
Dosage		
Total dose (Range)	27 - 45 g	1.98 - 2.03g
(mean)	36.0 g	2.0 g
Duration of Therapy (Days)		
Range	9 - 15	9 - 11
Mean	12.0	10.0

AZT - aztreonam
TOB - tobramycin
CLI - clindamycin

Table II(B)

Protocol 18554-38: Comparison of Aztreonam plus Clindamycin with Tobramycin plus clindamycin in the Treatment of Intra-abdominal Infection (Foreign Study)

Microbiologic Response

Gram-negative Pathogen	AZT + CLI Number eradicated/	TOB + CLI No. Treated
Single Pathogen:		
E. coli	1/1	1/1
P vulgaris	1/1	0/0
Ps. aeruginosa	1/1	0/0
Multiple pathogens:		
E. coli + Enterobacter sp.	1/1	0/0
E. coli + K. pneumoniae + Serratia sp.	0/0	1/1
Total	4/4	2/2

AZT - aztreonam
CLI - clindamycin
TOB - tobramycin

Protocol 18554-41 : Comparison of Aztreonam plus Clindamycin with Gentamicin plus Clindamycin in the Treatment of Gram-negative Obstetric and Gynecologic Infections

Domestic Study

This was a multicenter, single-blind, randomized study, in which the efficacy and safety of aztreonam was compared to that of gentamicin, an aminoglycoside, in the treatment of obstetric and gynecologic infections.

Four domestic principal investigators entered a total of 78 patients with endomyometritis into this study. Patients were randomly assigned to either the test or control drug in a ratio of 1:1. The investigators were not blinded. Forty patients were in the aztreonam group and thirty-eight in the gentamicin group. The demography of the two treatment groups was similar with respect to age, weight, and race, as presented in the applicant's Tables 2A and 2B (vol. 3.3:21040). The dosage regimens were: aztreonam - 1 to 2 g q 8 h, gentamicin - 80 to 140 mg q 8 h, intravenously. Patients in both groups received intravenous clindamycin at a dose of 600 mg q 6 to 8 hours. The duration of therapy ranged from 4 to 7 days.

As presented in Table I, the majority of patients who were entered into this study were excluded from the efficacy evaluation. In 87% of the 62 excluded patients, aerobic gram-negative pathogens were not isolated. The applicant pooled data from all investigators, irrespective of whether the investigator had evaluable patients in both treatment groups. Only one investigator (6435) had evaluable patients in both treatment groups. The demography of the evaluable patients in the two treatment groups was similar, as shown in Table I. The clinical signs and symptoms of endomyometritis were present in all patients, and single or multiple aerobic gram-negative pathogens were isolated, as shown in Table II. The criteria for cure (microbiologic and clinical) were eradication of the pathogens and/or resolution of symptoms and signs during and after completion of therapy. The applicant stated that microbiologic and clinical cures were achieved in all 13 patients (100%) in the aztreonam group, as compared to 6 (66.7%) of the 9 patients in the gentamicin group. The analyses of this study by this reviewer is presented in Table II. The number of patients in the aztreonam group is smaller than that presented by the applicant, nevertheless the results were comparable. Bacteriologic cure was seen in all 8 patients in the aztreonam group and in 5 of 8 patients in the gentamicin group. Superinfections occurred in 2 (5%) of the 40 aztreonam-treated patients and in 1 (3%) of the 38 gentamicin-treated patients. The causative microorganisms were S. faecalis in the aztreonam group and Enterobacter sp. in the gentamicin group. No other antibiotic therapy was given to these patients.

The safety of the two treatments were assessed in all patients who received the drugs. Adverse effects, possibly or probably related to drug therapy, were observed in 5% (2/40) of the aztreonam group and in 7.9% (3/38) of the gentamicin group. The adverse reactions were: one case each of diarrhea and pain at the infusion site in the aztreonam group; one case each of pruritic rash, diarrhea, and transient laboratory abnormalities (increased transaminases and alkaline phosphatase levels) in the gentamicin group.

Conclusions: Although a significant number of patients were entered into this multicenter, single-blind, randomized study, the number of patients that were evaluable for efficacy was quite small. Of the four domestic investigators who participated in this study, only one had evaluable patients in both treatment groups, and therefore, this study could be considered as a single-center study for the efficacy evaluation. The applicant concluded that the results of this study indicated that aztreonam is as safe and effective as gentamicin in the treatment of obstetric-gynecologic infections. This reviewer consideres such a conclusion to be rather premature, because of the small number of evaluable patients.

Table I

Protocol 18554-41 : Comparison of Aztreonam plus Clindamycin with Gentamicin plus Clindamycin in the Treatment of Gram-negative Obstetric and Gynecologic Infections

Number and (ID No.) of Principal Investigators: 4 (4889*, 6435, 7535*, 7653*)

	Treatment AZT + CLI	Treatment GEN + CLI
Total No. of Patients Entered:	40	38
Number of Patients Excluded from Efficacy Evaluation:	32	30
Reasons for Exclusion:		
Bacteriologic criteria not met (No gram-negative pathogens, inappropriate cultures)	25	29
Less than 5 days of therapy due to adverse reaction	1	0
Concurrent use of other antibiotic	1	0
Inadequate follow-up	1	0
Other (No evaluable patients in the test or control group)	4	1
Number of Patients Evaluable for Efficacy:	8	8
Demography and Other Characteristics of Evaluable Pts:		
Sex		
Female	8	8
Age (years)		
Range	17 - 37	15 - 3(
Mean	26.4	20.9
Race		
Caucasian	7	7
Black	1	1
Clinical Diagnosis:		
Endomyometritis	8	8
Dosage Regimen:	1 - 2 g q 8 h	1 - 1.5 mg/kg q 8 h
Total Dose: Range	21 - 30 g	0.95 - 2.1 g
Mean	27.4 g	1.7 g
Duration of Treatment (days):		
Range	4 - 5	3 - 7
Mean	4.6	5.4

*The investigator did not enter evaluable patients into both treatment groups.
AZT - aztreonam GEN - gentamicin CLI - clindamycin

Table II

Protocol 18554-41 : Comparison of Aztreonam plus Clindamycin with Gentamicin plus Clindamycin in the Treatment of Gram-negative Obstetric and Gynecologic Infections

Microbiological Respons

Gram-negative Pathogen	No. Eradicared/No. Treated AZT + CLI	GEN + CLI
Single Pathogen:		
E. coli	1/1	2/3
P. mirabilis	2/2	1/1
K. pneumoniae	2/2	0/1
Multiple Pathogens:		
E. coli + P. mirabilis	1/1	0/0
E. coli + K. pneumoniae + M morganii	0/0	1/1
E. aerogenes + K. pneumoniae	1/1	0/0
E. aerogenes + Ps. aeruginosa	1/1	0/0
K. pneumoniae + P. mirabilis + Ps. aeruginosa	0/0	0/1
Yersinia enterocolitica	0/0	1/1
Total	8/8	5/8

AZT - aztreonam
GEN - gentamicin
CLI - clindamycin

Uncontrolled (non-comparative) Clinical Studies

Protocol 18554-31: Aztreonam in the Treatment of Urinary Tract Infections due to Multidrug-resistant Microorganisms

This was a multicenter, single-drug study in which the efficacy and safety of aztreonam was evaluated in the treatment of UTI caused by gram-negative urinary pathogens resistant to aminopenicillins, cephalosporins, and/or aminoglycosides.

A total of 97 patients (96 domestic and 1 foreign) who met entrance criteria were entered into this study by 7 domestic and 1 foreign investigators. Three domestic investigators entered the majority of patients. Sixty-four patients were males and thirty-three were females. The age of patients raged from 20 to 100 years, with a mean of 64.8 years. The weight of patients ranged from 38.6 to 150 Kg, with a mean of 73 Kg. Forty-three patients were excluded from the efficacy evaluation by this reviewer, for reasons listed in Table I (A). The demographics and other characteristics of the evaluable patients are also presented in the table. Forty-five (83%) of the 54 evaluable patients had complicated UTI. In twenty-one of the patients (15 males and 6 females) with complicated UTI, urosurgical procedures were performed during therapy. Thirty-five (64.8%) of the evaluable patients were male. Pseudomonas aeruginosa was the predominant uropathogen (57%) in this study population. The dosage of aztreonam ranged from 0.5 to 2 g every 8 to 12 hours. The majority of patients were treated with a dosage regimen of 1 g q 8 h. The drug was administered intravenously in 47, intramuscularly in 5, and both intravenously and intramuscularly in the remaining 2 patients.
The clinical and laboratory monitoring of patients in this study was similar to that in preceding controlled UTI studies.

The applicant reported an overall bacteriologic cure rate of 88% (60/68), and a favorable clinical response rate of 96%, at 5-9 days after completion of therapy (vol. 3.3 : 807 & 819). It was noteworthy that bacteriologic cure for P. aeruginosa was reported to be 87% (34/39). This reviewer's evaluation of bacteriologic responses at a later follow-up, 4-6 weeks post-therapy, is presented in Table II(A-B) The overall bacteriologic cure rates were 82.5% (33/40) for complicated UTI and 77.8 % (7/9) for uncomplicated UTI. The bacteriologic cure rate for P. aeruginosa UTI was 71% (17/24) in complicated UTI, and 80% (4/5) in uncomplicated UTI. The results for complicated UTI appeared to be more favorable than those in the preceding controlled UTI studies, particularly for UTI caused by P. aeruginosa. This might be attributable to an unusually high cure rate seen in one major investigator's study (0121). Reinfection occurred in 17.8% (8/45). All microorganisms causing reinfections, except for S. faecalis, were susceptible to aztreonam. Superinfection occurred in 11% of the 97 patients who were treated with aztreonam. The superinfection rate for the patients who were evaluable for efficacy remained the same, 11% (5/45). The causative organism was S. faecalis in 10, and S. aureus and S. faecalis in 1. Four of the eleven patients with superinfection were treated with other antimicrobial agents.

The safety of aztreonam was assessed in all patients who received the drug in this uncontrolled study. Clinical adverse effects which were possibly or probably attributable to aztreonam were reported in 2 patients (2%); those were one case each of rash and phlebitis. The patient who developed rash also received ampicillin concurrently. Laboratory abnormalities were noted in 5 patients (5%); those were eosinophilia in 1, increased AST(SGOT) and/or ALT(SGPT) in 4, and increased alkaline phosphatase levels in 1. In none of these patients was the drug discontinued because of the adverse effects. Death occurred in 5 patients, but these were not attributed to drug therapy.

Conclusions: This multicenter study of aztreonam in hospitalized patients with complicated and/or recurrent UTI, caused by gram-negative uropathogens resistant to amidopenicillins and cephalosporins, indicated that aztreonam is relatively effective and safe in this study population. The results of this uncontrolled study, however, appeared to be more favorable to aztreonam than those of the other multicenter, randomized, controlled UTI studies. It should be recognized that the interpretation of this data is limited, due to the design of this study.

Table I (A)

Protocol 18554-31: Aztreonam in the Treatment of Urinary Tract Infection due to Multidrug-resistant Microorganism

Domestic Study

Investigators' Number: 0121; 4701; 5023*; 6208, 6215; 6218, 6494*

	Aztreonam
Total Number of Patients Entered:	97
No. of Patients Not Evaluable for Efficacy	43
Reasons:	
Improper or negative pretreatment culture	11
Inappropriate follow-up	18
5 days of therapy	3
Concurrent antimicrobial therapy	5
Clinical diagnosis other than UTI	6
No. of Patients Evaluable for Efficacy*	54
Demographic Characteristics:	
Sex	
Female	19
Male	35
Age (Years)	
Range	20 - 88
Mean	66.8
Race	
Black	31
Caucasian	23
Clinical Diagnosis	
UTI (unspecified)	31
Pyelonephritis	12
Cystitis	9
Other (asymptomatic bacteriuria)	2
Complicated UTI	45
Uncomplicated UTI	9
Route of Administration:	
IM	47
IV + IM	2
IM	5
Duration of Treatment (days)	
Range	5 - 16
Mean	8.3

*The investigator did not enter patients who are evaluable for efficacy.
**Patients who had appropriate follow-up up to 4-6 weeks after completion of therapy.

Table II (A)

Protocol 18554-31: Aztreonam in the Treatment of Urinary Tract Infections due to Multidrug-resistant Gram-negative Microorganism

Domestic Study

Complicated and Uncomplicated Urinary Tract Infection
Bacteriologic Response *

Pathogen	Complicated UTI E	P	RL	RI	SI	Uncomplicated UTI E	P	RL	RI	SI
Single pathogen:										
P. aeruginosa	16/22	3	0	7(5-L)	1	4/5	1	0	0	0
E. coli	4/6	1	0	1(L)	1	1/2	1	0	0	0
P. rettgeri	4/4	0	0	0	0	1/1	0	0	0	0
E. cloacae	3/3	0	0	0	0	1/1	0	0	0	0
K. pneumoniae	1/2	1**	0	0	1**	0/0	-	-	-	-
E. aerogenes	2/2	0	0	0	0	0/0	-	-	-	-
M. morganii	2/2	0	0	0	0	0/0	-	-	-	-
C. freundii	1/1**	0	0	0	1**	0/0	-	-	-	-
Multiple pathogens:										
P. aeruginosa + P. stuartii	0/3	2	0	0	1	0/0	-	-	-	-
Total	33/45 (73%)	7	0	8(E-2) (6-L)	5	7/9	2	0	0	0

* At 4-6 weeks after completion of therapy ** The same patients
E- eradication; number eradicated/number treated
P - persistence
RL - relapse
RI - reinfection
SI - superinfection
(E) - Reinfection at 5-9 days after completion of therapy
(L) - Reinfection at 4-6 weeks after completion of therapy

Table II (B)

Protocol 18554-31: Aztreonam in the Treatment of Urinary Tract Infections due to Multidrug-resistant Gram-negative Microorganism

Domestic Study

Urinary Tract Infection (Complicated + Uncomplicated)
Bacteriologic Cure (eradication of original pathogen)*

UTI	Number Cured/Number Treated** (cure rate)
Complicated	33/40 (82.5%)
Uncomplicated	7/9 (77.8%)
Total	40/49 (81.6%)

Pseudomonas aeruginosa:

Complicated UTI	17/24 (70.8%)
Uncomplicated UTI	4/5
Total	21/29 (72.4%)

*At 4 - 6 weeks after completion of therapy
**Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections due to Aerobic Gram-negative Organisms

This was a multicenter, open single-drug study of aztreonam in the treatment of hospitalized patients with a wide variety of infections caused by aerobic gram-negative microorganisms. The infections studied were those involving the bone and joints, the skin and skin structures, intra-abdominal, obstetric and gynecologic organs, the urinary and lower respiratory tracts, and septicemia.

The initial NDA submission by the applicant stated that a total of 105 investigators, 59 domestic and 46 foreign, participated in this study, and that a total of 963 patients, 514 domestic and 449 foreign, were treated with aztreonam. Five-hundred fourteen patients, 322 domestic and 192 foreign, were unevaluable for efficacy. The major reasons for exclusions were a failure to isolate gram-negative pathogens susceptible to the drug prior to therapy and an appropriate post-therapy follow-up. A total of 473 sites of infection were treated among the 449 evaluable patients. An NDA amendment (case summary tables according to source, domestic or foreign, and infection site) submitted on October 7, 1985 provided an additional 200 patients (123 domestic and 77 foreign) who were entered into this open study. Some patients had infections involving multiple sites and, therefore, the number of infection sites treated were larger than the total number of patients.

The distribution of infection sites among the evaluable patients is shown in the following table.

Infection Site	SQ	MO
Urinary Tract	106	109 (80-D, 29-F)
Skin/skin structures	136	138 (104-D; 34-F)
Lower Respiratory Tract	119	117 (80-D; 37-F)
Septicemia	63	123 (80-D; 43-F)
Intra-abdominal	29	56 (37-D, 19-F)
Bone and Joint	12	29 (25-D, 4-F)
Obstetric/Gynecologic	8	23 (22-D; 1-F)
Total	473	595 (428-D;167-F)

SQ - Squibb & Sons : The numbers in this column are derived from the applicant's table (vol. 3.3, p.-21077).
MO - Medical Officer's tabulation based on data submitted on 10/7/85.
D - Domestic study
F - Foreign study

The evaluation of the microbiological and clinical results by this reviewer included those additional cases submitted recently by the applicant. The evaluation of the data on each indication is as follows:

Bone and Joint infections:

Twenty-three principal investigators, 19 domestic and four foreign, treated 46 hospitalized patients (41 domestic and 5 foreign) with bone and joint infections. Two-thirds of the patients had osteomyelitis, and the remaining had septic arthritis. The majority of the patients were male, as shown in Table I.
The patients with clinical symptoms and signs compatible with the disease, i.e., draining sinus, local swelling, local tenderness, and limitation of motion, were entered into this study. The diagnoses were comfirmed by radiographic findings and/or cultures of the bone biopsy or aspirate specimens. Susceptibility testing was done using the disc or tube dilution method.

The treatment regimen consisted of aztreonam 2 g q 6-8 h IV for 2-6 weeks. The use of other antibiotics was allowed, provided that the antibiotics possessed little activity against gram-negative pathogens, but were known to be active against anaerobes and gram-positive pathogens.

Of the forty-six patients treated, 17 patients (16 domestic and 1 foreign) were excluded from the efficacy evaluation. The reasons for exclusions were inappropriate pretreatment culture, surgical procedure (amputation), inadequate duration of therapy, and concurrent use of antibiotics which are active against gram-negative pathogens. Of the twenty-nine patients evaluable for efficacy, 25 were from domestic studies, and 4 were from foreign studies.

As shown in Table I, 19 patients had osteomyelitis, 11 acute and 8 chronic, and 10 had septic arthritis. In all except two foreign patients the drug was administered intravenously. Seven patients were also treated concurrently with clindamycin, and four with vancomycin or nafcillin, intravenously. The duration of therapy ranged from 27 to 50 days for osteomyelitis, and 12 to 43 days for septic arthritis. Surgical intervention (debridement) was done during therapy in 2 patients. For the efficacy evaluation of osteomyelitis, those patients who received at least 4 weeks of therapy were included. Monitoring of patients included daily clinical observation, periodic laboratory tests (hemogram, urinalyses, and blood chemistsries), and repeated cultures of specimen when it was obtainable. The applicant's criteria for cure used for osteomyelitis were similar to those used in the evaluation of Other indications. In the majority of the study population, the duration of post-therapy follow-up was rather short, less than 1 week after completion of therapy. In only 2 patients with chronic osteomyelitis, was the post-therapy follow-up longer than 4 weeks. This reviewer considered that the duration of the post-therapy follow-up was inadequate to ascertain the clinical outcome (cure) of the disease, especially in patients with osteomyelitis. The reviewer, therefore, used the term 'clinical improvement' rather than "clinical cure". In the evaluation of microbilogic response, eradication of pathogens was assumed when clinical improvement was seen. The assumed microbiologic eradication was seen in 15 of the 17 patients (88.2%) with osteomyelitis, and in 6 of the 8 patients (75%) with septic arthritis, as shown in Table II. In one patient with chronic osteomyelitis caused by Pseudomonas aeruginosa, which was not eradicated, a change in susceptibility to aztreonam was demonstrated by an increase in the MIC. Clinical improvement was seen in all patients. The applicant reported earlier that all 12 evaluable patients with infections of the bones and joints were microbiologically cured.
Clinical cure was reported by the applicant in 10 patients, with a partial response in the remaining 2 patients.

The safety of aztreonam in the treatment of the infections of the bones or joints was evaluated in all 46 patients who were treated with the drug. Adverse reactions possibly or probably related to the drug were reported in 9 patients (19.6%), 8 domestic and 1 foreign. The reactions observed were as follows:

	No. of Patients with AR/No. of Patients Treated	
	Domestic Study	Foreign Study
Osteomyelitis	8/31	1/2
Clinical:	6	0
Rash	1	0
Phlebitis	2	0
Halitosis/taste of drug	2	0
Pruritus	1	0
Laboratory:	3	1
Elevated ALT(SGPT)/AST(SGOT)	2	0
Eosinophilia	1	1
Septic arthritis	0/10	0/2
Total	8/41	1/4

Conclusions: The overall results of this open study of aztreonam appeared favorable in the treatment of the patients with acute osteomyelitis and septic arthtitis caused by aerobic gram-negative microorganisms (Pseudomonas aeruginosa, Escherichia coli, and Proteus mirabilis). However, the number of patients studied was small, and the duration of post-therapy follow-up was inadequate to ascertain efficacy of aztreonam in the study population. The favorable therapeutic results seen in this uncontrolled study should, therefore, be confirmed by adequate well controlled clinical trials of this drug compared with other antibiotics approved for this indication.

Protocol 18554-16: Evaluation of / ...d in the Treatment of Serious Infections (Bone and Joint) due to Gram-negative Organisms

Table I

	Domestic	Foreign
Number of Investigators	19	4
Investigators' ID Number:	2891;4232;6067;6173; 6207;6209;6210;6226; 6243;6245;6411;6437; 7510;7512;7516;6424, 7534;7556;7665	6228,6310,6430;6452
Number of Patients Entered	41	5
Demography:		
Sex		
Female	9	0
Male	32	5
Age		
Range	19 - 75	21 - 82
Mean	49.5	43.8
Race		
Black	13	0
Caucasian	25	4
Other	3	1
Diagnosis		
Osteomyelitis	31	3
Septic arthritis	10	2
Number of Patients Excluded frome Efficacy Evaluation	16	1
Number of Patients Evaluable for Efficacy	25	4
Diagnosis:		
Acute ostemyelitis	11	0
Chronic osteomyelitis	6	2
Septic arthritis	8	2
Dosage Regimen:	2 g q 6 - 8 h IV	2 g q 8 h IV or IM
Duration of Therapy (days):		
for Osteomyelitis:		
Range/(mean)	27 - 50 (40.4)	42 - 43
for Septic arthritis	12 - 42 (24.6)	14 - 43

Table II

Bone and Joint Infections

Microbiologic Response

Infection/Pathogen	Number Eradicated*/Number Treated	
	Domestic	Foreign
Osteomyelitis:		
Pseudomonas aeruginosa	5/6	1/1
Enterobacter cloacae	3/3	-
Proteus mirabilis	3/3	-
Escherichia coli	2/2	1/1
Serratia marcescens	1/1	-
M. morganii + P. vulgaris	1/1	-
E. coli + P. aeruginosa**	0/1	-
Total	15/17 (88.2%)	2/2
Septic Arthritis:		
E. coli	1/2	-
P. aeruginosa	2/2	1/1
Enterobacter aerogenes	0/1	-
S. marcescens	1/1	1/1
Haemophilus	1/1	-
P. aeruginosa + K. pneumoniae	1/1	-
Total	6/8 (75%)	2/2
Acute osteomyelitis	10/11 (90.9%)	-
Chronic osteomyelitis	5/6	2/2

*Eradication was assumed at the 1-4 week post-therapy follow-up.
** The microorganism was not eradicated.

Intra-abdominal Infections

Forty principal investigators, 25 domestic and 15 foreign, participated in this multicenter, non-comparative study of aztreonam in the treatment of intra-abdominal infections caused by aerobic gram-negative microorganisms. A total of 80 patients, 46 domestic and 34 foreign, were treated. The demographic characteristics of 56 evaluable patients, 37 domestic and 19 foreign, are presented in Table I. The most common clinical diagnoses were intra-abdominal abscess and peritonitis. Three of the 37 domestic patients also had septicemia. The dosage regimen of aztreonam was similar to that used in the multicenter controlled study on the same indication. The drug was administered intravenously in all of the domestic study patients and in 6 of the 19 foreign study patients. The duration of therapy ranged from 5 to 44 days. Necessary surgical procedures were performed in 29% of patients, and antimicrobials effective for anaerobes, clindamycin or metronidazole were administered concurrently in 63% of patients. The most common aerobic gram-negative pathogens isolated were E. coli, P. aeruginosa, and K. pneumoniae. Polymicrobial infection occurred in 49% (18/37) of the domestic study patients and in 16% (3/19) of the foreign study patients. The overall microbiological response w less favorable in the domestic study population, as shown in Tables II and I.I. This might be attributable to the complexity of polymicrobial infections. The overall eradication rates were 90% (27/30) for E. coli, 75% (12/16) for P. aeruginosa, and 100 % (12/12) for K. pneumoniae. Favorable clinical responses (clinical cure or improvement) were seen in 89% of the domestic study population and in 100 % of the foreign study population. The overall results seen in this non-comparative study were similar to those seen in limited number of patients in the multicenter, comparative study of intra-abdominal infections which was reviewed earlier. Superinfection occurred in 2 patients in domestic studies. One was due to enterococci and the other to Enterobacter aerogenes. Both patients were treated with other antibiotics.

The safety of aztreonam therapy was assessed in 80 patients who received the drug. Adverse effects which were possibly or probably related to the drug were observed in 7 patients, 6 domestic and 1 foreign. These were rashes in 2 patients, and one case each of dizziness and somnolence, candidiasis, pain at the injection site, thrombocytopenia and bleeding at the incision site, and elevated alkaline phosphatase level. Deaths occurred in 12 patients, 11 domestic and 1 foreign. None of the deaths was attributed to drug therapy.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious (Intra-abdominal) Infections due to Gram-negative Organisms

Table I

	Domestic	Foreign	Total
Number of Investigators	25	15	40
Number of Patients Entered	46	34	80
Number of Patients Excluded from Efficacy Evaluation	9	15	24
Number of Patients Evaluable for Efficacy	37	19	56
Demographic Characteristics:			
Sex			
Female	13	11	24
Male	24	8	32
Age			
Range	22 - 85	8 - 76	
Mean	58.8	51.3	
Race			
Caucasian	32	19	51
Black	4	0	4
Other(not stated)	1	0	1
Clinical Diagnosis:			
Intra-abdominal abscess (including retroperitoneal,	18	9	27
Cholangitis/cholecystitis	8	8	16
Other intra-abdominal infections	9	-	9
	2	2	4
Dosage Regimen:			
IV 1-2 g q 6-8 h	37	13	50
IM 1-2 g q 8 h	(	6	6
Duration of Therapy (days):			
Range	5 - 20	8 - 44	
Mean	11.9	17.4	
Concurrent antibiotics used (for anaerobic or gram-positive microorganisms)	26	9	35
Surgical Intervention	11	6	17

Table II

Intra-abdominal Infections

Microbiologic Response

Pathogen	No. Eradicated/No of Patients Treated		
	Domestic	Foreign	Total
Single Pathogen:			
Escherichia coli	7/8	7/7	14/15 (93%)
Pseudomonas aeruginosa	5/8	4/4	9/12 (75%)
Klebsiella pneumoniae	2/2	1/1	3/3
Klebsiella sp.	0/1	-	0/1
Serratia marcescens	-	1/1	1/1
E. cloacae	-	1/1	1/1
Citrobacter sp.	-	1/1	1/1
Aeromonas hydrophila	-	1/1	1/1
Multiple Pathogens:			
E. coli + C. freundii	1/1	-	1/1
E. coli + K. pneumoniae	3/3	1/1	4/4
E. coli* + E. cloacae	1/2	-	1/2
E. coli + E. aerogenes	0/1	-	0/1
E. coli + P. aeruginosa	2/2	-	2/2
E. coli + Pseudomonas sp.	1/1	-	1/1
E. coli + S. marcescens	-	1/1	1/1
E. aerogenes + P. aeruginosa	0/1	-	0/1
E. cloacae* + S. marcescens	0/1	-	0/1
K. pneumoniae + C.freundii	1/1	-	1/1
K. pneumoniae + K. oxytoca	1/1	-	1/1
K. oxytoca + P. aeruginosa*	0/1	-	0/1
[redacted]	1/1	[redacted]	1/1
+ E. cloacae			
E. coli + K. pneumoniae + P. mirabilis	-	1/1	1/1
E. coli + S. liquefaciens + S. marcescens*	0/1	-	0/1
Total	24/37 (64.8%)	19/19 (100%)	45/56 (80.4%)

*The microorganism not eradicated.

Table III

Intra-abdominal Infections

Microbiologic Response

Pathogen	No. of Isolates Eradicated/No of Patients Treated		
	Domestic	Foreign	Total
Escherichia coli	17/20 (85%)	10/10	27/30 (90%)
Pseudomonas aeruginosa	8/12 (67%)	4/4	12/16 (75%)
Klebsiella pneumoniae	9/9	3/3	12/12 (100%)
E. cloacae	2/3	1/1	3/4
Serratia marcescens	1/2	2/2	3/4
Klebsiella oxytoca	2/2	-	2/2
Citrobacter freundii	2/2	-	2/2
Klebsiella sp.	0/1	-	0/1
Pseudomonas sp.	1/1	-	1/1
Citrobacter sp.	-	1/1	1/1
Aeromonas hydrophila	-	1/1	1/1
Aeromonas sp.	1/1	-	1/1
P. mirabilis	-	1/1	1/1
S. liquefasciens	1/1	-	1/1
Total	44/54 (81.4%)	23/23 (100%)	67/77 (87.0%)

Obstetric and Gynecologic Infections

Five principal investigators, 4 domestic and 1 foreign, studied a total of 26 patients who were hospitalized for the treatment of OB/GYN infections. Twenty-two of the 26 patients were evaluable for efficacy. The demography of evaluable patients is presented in Table I. The most common pathogens isolated were N. gonorrhoeae and E. coli.

A majority of the patients received a 5-day course of aztreonam therapy (1-2 g q 8 h), intravenously. Clindamycin was concurrently used to cover obligate anaerobes in 17 patients. As noted in Table II, the overall microbiological response was favorable; The eradication rates for Enterobacteriaceae and N. gonorrhoeae were 89% and 100 %, respectively. The clinical response was reported to be favorable in all patients. The total number of patients treated, however, was small and the duration of post-therapy follow-up was rather short in more than one-half of the patients. No clinical adverse reactions were reported in this study, but laboratory abnormalities were noted in 3 PID patients, namely, eosinophilia in 2, and prolonged prothrombin time in 1.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (Obstetric and Gynecologic) due to Gram-negative Organisms

Table I

	Domestic	Foreign
Number of Investigators	4	1
Investigators' ID Number:	6437,7510;7533,7723;	76630
Number of Patients Entered	25	1
Number of Patients Not evaluable for Efficacy	3	0
Reasons:		
Susceptibility test not done	2	0
Inadequate follow-up	1	0
Number of Evaluable Patients for Efficacy:	22	1
Demography:		
Sex		
Female	22	1
Age		
Range	16 - 65	24
Mean	24.3	-
Race		
Black	15	0
Caucasian	7	0
Diagnosis		
Endometritis	5	1
Pelvic abscess	3	0
Pelvic inflammatory disease(PID)	14	0
Dosage Regimen(IV):	1-2 g q 8 h	1-2g q 8-12 h
Duration of Therapy (days):		
Range	5 - 11	10
Mean	5.9	-

Table II

Obstetric and Gynecologic Infections

Microbiologic Response

Pathogen	Number Eradicated/Number Treated	
	Domestic	Foreign
Escherichia coli	6/7	-
Proteus mirabilis	1/1	-
E. coli + E. aerogenes	1/1	1/1
Total	8/9	1/1
Neisseria gonorrhoeae*	13/13	-

* PID patients.

Septicemia

Sixty four investigators, 33 domestic and 31 foreign, entered a total of 197 patients, 111 domestic and 86 foreign, with septicemia (primary and secondary) into this uncontrolled study. The number of patients was larger than that presented in the initial NDA submission, since this review included additional patients on whom case reports were submitted by the applicant in October 1985. The demographic characteristics of these patients are presented in Table I. The majority of patients had secondary septicemia. Urinary and lower respiratory tracts were the most common primary sites of infection. One hundred twenty-three patients, 80 domestic and 43 foreign, were evaluable for efficacy. The evaluable patients had at least 2 blood cultures positive for aztreonam-susceptible pathogens obtained within 48 hours prior to initiation of therapy. Escherichia coli, Klebsiella pneumoniae, and Pseudomonas aeruginosa were the most common gram-negative pathogens isolated. Susceptibility of clinical isolates was demonstrated by the disc-diffusion or agar dilution method. The majority of patients had infections caused by single gram-negative pathogens.

The dosage regimen in the domestic and foreign studies ranged from 1 to 2 g every 6 to 12 hours intravenously. Mean duration of therapy was 12 days.

The overall microbiologic responses seen in the domestic and foreign study populations were similar, and were very favorable, as shown in Tables II and III. The overall eradication rates were 98% (57/58) for E. coli, 90% (13/15) for P. aeruginosa, and 87% (13/15) for K. pneumoniae. Although the numbers of other pathogens, Proteus mirabilis, Serratia marcescens, and Enterobacter species were smaller (less than 10 cases each), all of these pathogens were eradicated. Reinfections caused by K. pneumoniae and Clostridium perfringens were reported in one case each. Superinfections or colonizations were due to enterococci (S. faecalis) in 4, S. aureus in 2, and S. epidermidis in 1. A few of these patients were treated with other antibiotics.
The overall clinical responseswere also favorable, although a few critically ill patients died within 48 hours of starting therapy. The favorable clinical responses (clinical improvement or partial responses) were seen in 94% (116/123). These results were similar to those reported by the appplicant in its earlier analyses of a smaller number of patients.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Seri .s Infectious (Septicemia) due to Gram-negative Organ.sms

Table I

	Domestic	Foreign	Total
Number of Investigators	33	31	64
Number of Patients Entered	111	86	197
Demography:			
Sex			
Female	54	27	81
Male	57	59	116
Age			
Range	19 - 97	8 - 87	
Mean	62.4	59.2	
Race			
Black	31	1	32
Caucasian	79	80	159
Other	1	5	6
Primary site of infections:			
Urinary tract infection	48	36	84
Bone and Joint infection	2	0	2
Intra-abdominal inf.	6	12	18
Lower respiratory tract inf.	5	3	8
Obstetric/gynecologic inf.	1	0	1
Skin structure inf.	3	5	8
Number of Patients Excluded frome Efficacy Evaluation	31	43	74
Number of Patients Evaluable for Efficacy	80	43	123
Dosage Regimen(IV):	1-2 g q 6 - 8 h	1-2 g q 8 -12 h	
Duration of Therapy (days):			
Range	2 - 37	2 - 41	
Mean	12.7	11.7	

Table II

Septicemia

Microbiologic Response

Pathogen	Number Eradicated/Number Treated		
	Domestic	Foreign	Total
Single Pathogen:			
Escherichia coli	32/32	23/24	55/56 (98%)
Klebsiella pneumoniae	5/6	6/6	11/12 (92%)
Pseudomonas aeruginosa	5/6	2/2	7/8
Proteus mirabilis	5/5	-	5/5
Serratia marcescens	5/5	3/3	8/8
Enterobacter aerogenes	5/5	1/1	6/6
E. cloacae	2/2	2/2	4/4
Citrobacter diversus	2/2	-	2/2
Haemophilus influenzae	2/2	1/1	3/3
Providencia stuartii	2/2	1/1	3/3
Citrobacter freundii	1/1	-	1/1
Enterobacter agglomerans	1/1	-	1/1
Haemophilus sp.	1/1	-	1/1
K. oxytoca	-	1/1	1/1
Morganella morganii	1/1	-	1/1
P. vulgaris	1/1	-	1/1
Salmonella typhi	1/1	-	1/1
Salmonella enteritidis	1/1	1/1	2/2
Multiple Pathogens			
E. coli + P. mirabilis	1/1	-	1/1
E. coli + P. stuartii	1/1	-	1/1
C. freundii + P. aeruginosa	1/1	-	1/1
K. pneumoniae + K. oxytoca	1/1	-	1/1
K. pneumoniae + P. aeruginosa	1/1	-	1/1
K. pneumoniae + S. liquefaciens	0/1	-	0/1
S. typhimurium + S. marcescens	-	1/1	1/1
Total	77/80 (96.3%)	42/43 (97.7%)	119/123 (96.7%)

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Table III

Septicemia

Microbiologic Response

Pathogen	Number of Isolates Eradicated/Number Treated		
	Domestic	Foreign	Total
Escherichia coli	34/34	23/24	57/58 (98%)
Klebsiella pneumoniae	7/9	6/6	13/15 (87%)
Pseudomonas aeruginosa	7/8	2/2	9/10 (90%)
Proteus mirabilis	6/6	-	6/6
Serratia marcescens	5/5	4/4	9/9
Enterobacter aerogenes	5/5	1/1	6/6
E. cloacae	2/2	2/2	4/4
Citrobacter diversus	2/2	-	2/2
Haemophilus influenzae	2/2	1/1	3/3
Providencia stuartii	3/3	1/1	4/4
Citrobacter freundii	2/2	-	2/2
Enterobacter agglomerans	1/1	-	1/1
Haemophilus sp.	1/1	-	1/1
K. oxytoca	1/1	1/1	2/2
Morganella morganii	1/1	-	1/1
vulgaris	1/1	-	1/1
Salmonella enteritidis	1/1	1/1	2/2
Salmonella typhi	1/1	-	1/1
S. typhimurium	-	1/1	1/1
Total	87/91 (95.6%)	43/44 (97.7%)	130/135 (96.3%)

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Skin and Skin structure Infections

Forty-five investigators, 36 domestic and 9 foreign, entered a total of 161 patients, 121 domestic and 40 foreign, with infections of the skin and skin structures into this single-drug study. Of these, 138 patients, 104 domestic and 34 foreign, were evaluable for efficacy. The demographic characteristics of the evaluable patients are presented in Table I. The most common clinical diagnoses were cellulitis and wound infection in the domestic study population, and wound and burn infections were in the foreign study population. Five domestic patients had multi-site infections. Pseudomonas aeruginosa, Proteus mirabilis, E. coli, Klebsiella-Enterobacter species, and Serratia marcescens were the most common pathogens isolated in the study population. All of the clinical isolates were susceptible to aztreonam. The dosage regimen ranged from 0.5 g q 4-12 h to 2 g q 6-12 h. The regimens most often used were 1 g q 8 h and 2 g q 6-8 h. The majority of patients, 93 domestic and 14 foreign, received the drug intravenously. The duration of therapy ranged from 5 to 53 days with a mean of 14 days. Sixty patients, 55 domestic and 5 foreign, received concurrently other antimicrobial agents to cover anaerobes and gram-positive microoganisms. Surgical intervention, such as incision and drainage (I&D) and debridement were done in one-third of the patients.

The microbiologic response seen in the domestic and foreign studies was similar, as shown in Tables II and III. The eradication rate for polymicrobial infections was lower than that for single-pathogen infections. The overall eradication rates were 66% (38/58) for P. aeruginosa, 90% (27/30) for P. mirabilis, 85% (23/27) for E. coli, 92% (11/12) for S. marcescens, 86% (12/14) for K pneumoniae, and 93% (13/14) for Enterobacter species. P. aeruginosa isolates from three patients with microbiologic failure were shown to be resistant by the disk-diffusion or agar dilution method. Reinfection caused by P. aeruginosa occurred in 1 domestic patient. This isolate was reported to be intermediately susceptible to aztreonam. The most common isolates for superinfections (or colonizations) were S. aureus, Pseudomonas species, including P. aeruginosa and P. maltophilia, and other Enterobactericeae. The incidence of favorable clinical responses (clinical improvement and partial responses) seen in the domestic and foreign study population was similar. Overall favorable clinical responses were achieved in 93% (129/138). The rates for microbiologic eradication and favorable clinical response as determined by this reviewer were similar to those reported by the applicant.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (Skin/Skin Structures) due to Gram-negative Organisms

Table I

	Domestic	Foreign	Total
Number of Investigators	36	9	45
Number of Patients Treated	121	40	161
Number of Patients Evaluable for Efficacy	104	34	138
Demographic Characteristics:			
Sex			
Female	46	12	58
Male	58	22	80
Age			
Range	7 - 96	12 - 85	
Mean	59	55	
Race			
Caucasian	70	34	104
Black	32	-	32
Other	2	-	2
Clinical Diagnosis:			
Cellulitis	48	1	49
Wound infection	31	6	37
Skin infection	7	12	19
Abscess	9	3	12
Decubitus ulcer/ulceration	8	1	9
Burn infection	1	11	12
Dosage Regimen(range)	0.5 g q 4-6 h to 1 - 2 g q 6-12 h	0.5 g q 12 h to 1 - 2 g q 6-12 h	
Route of Administration:			
IV	93	14	107
IM	5	17	22
IV + IM	6	3	9
Duration of Therapy (days):			
Range	5 - 53	25 - 42	
Mean	14	15	
Concurrent antibiotics used: (for anaerobic and/or gram-positive microorganisms)	55	5	60
Surgical interventions: (I & D or debridement)	39	5	44

Table II

Skin and Skin Structure Infections

Microbiologic Response

Pathogen	No. Eradicated/No. of Patients Treated		
	Domestic	Foreign	Total
Single Pathogen:			
Pseudomonas aeruginosa	17/29 (59%)	11/14 (79%)	28/43 (65 %)
Proteus mirabilis	9/9	2/2	11/11 (100%)
Serratia marcescens	8/8	1/1	9/9 (100%)
Klebsiella pneumoniae	7/7	-	7/7
Escherichia coli	6/6	2/3	8/9
Enterobacter cloacae	2/3	2/2	4/5
E. aerogenes/Enterobacter sp.	2/2	-	2/2
Citrobacter freundii	1/1	1/1	2/2
K. oxytoca	1/1	-	1/1
Morganella morganii	1/1	-	1/1
Salmonella enteritidis	1/1	-	1/1
Citrobacter diversus	1/1	-	1/1
Enterobacter agglomerans	-	1/1	1/1
K. ozaenae	-	2/2	2/2
Providencia rettgeri	-	1/1	1/1
Pseudomonas cepacia	-	1/1	1/1
P. mallei	-	1/1	1/1
P. putida	-	1/1	1/1
Serratia liquefaciens	-	1/1	1/1
S. rubidaea	-	1/1	1/1
Total	56/69 (81%)	28/32 (88%)	84/101 (83%)
Multiple Pathogens:			
E. coli + P. mirabilis + P. aeruginosa	4/8* - 2/3	- 2/3	4/8 E. coli
E. coli + Pseudomonas sp.	1/1	-	1/1
C. freundii + K. oxytoca	1/1	-	1/1
E. aerogenes + Enterobacter sp.	1/1	-	1/1
E. aerogenes + P. aeruginosa	1/1	-	1/1
E. aerogenes + Pseudomonas sp.	1/1		1/1
K. pneumoniae + E. agglomerans	1/1	-	1/1
K. pneumoniae* + P. mirabilis	0/1	-	0/1
P. mirabilis + P. aeruginosa**	1/2	-	1/2
P. mirabilis + S. marcescens	1/1	-	1/1
P. aeruginosa + S. marcescens	1/2	-	1/2
P. aeruginosa** + M. morganii	-	0/1	0/1
P. mallei + Aeromonas sp.	-	1/1	1/1

Table II (continued)

Skin and Skin Structure Infections

Multiple Pathogens:			
C. freundii* + E. coli + K. pneumoniae	0/1	-	0/1
C. freundii + E. cloacae + K. oxytoca	1/1	-	1/1
E. coli + E. cloacae + K. pneumoniae	1/1	-	1/1
E. coli + P. mirabilis + E. aerogenes	1/1	-	1/1
E. coli + P. mirabilis + P. aeruginosa	1/1	-	1/1
E. coli + P. vulgaris + P. aeruginosa	1/1	-	1/1
E. coli + Citrobacter sp. + P. mirabilis + P. aeruginosa	1/1	-	1/1
K. pneumoniae + P. mirabilis + Morganella sp.	1/1	-	1/1
K. pneumoniae + P. mirabilis + P. aeuriginosa	1/1	-	1/1
P. mirabilis + P. vulgaris + Alcaligenes sp.	1/1	-	1/1
P. mirabilis + P. aeruginosa** + P. stuartii	0/1	-	1/1
Total	24/35 (69%)	1/2	25/37 (68%)
Total (single + multiple)	80/104 (76.9%)	29/34 (85.3%)	109/138 (78.9%)

*In the remaining 4 patients, P. aeruginosa was not eradicated in 3, and E. coli was not eradicated in 1.
** The microorganism was not eradicated.

Table III

Skin and Skin Structure Infection

Microbiologic Response

Pathogen	No. of Isolates Eradicated/Number Treated		
	Domestic	Foreign	Total
Pseudomonas aeruginosa	27/43 (63%)	11/15 (73%)	38/58 (66%)
Pseudomonas species	2/2	-	2/2
Proteus mirabilis	25/28 (89%)	2/2	27/30 (90%)
Escherichia coli	21/24 (88%)	2/3	23/27 (85%)
Serratia marcescens	10/11 (91%)	1/1	11/12 (92%)
Klebsiella pneumoniae	12/14 (86%)	-	12/14 (86%)
Enterobacter sp.(E. cloacae; E. aerogenes; E. agglomerans)	11/12 (92%)	3/3	14/15 (93%)
Citrobacter sp. (C. freundii/C. diversus)	5/7	-	5/7
K. oxytoca	3/3	-	3/3
Proteus vulgaris	2/2	-	2/2
Morganella morganii	1/1	1/1	2/2
P. stuartii	1/1	-	1/1
S. enteritidis	1/1	-	1/1
Alcaligenes sp.	1/1	-	1/1
Aeromonas sp.	-	1/1	1/1
K. ozaenae	-	2/2	2/2
Providencia rettgeri	-	1/1	1/1
Pseudomonas cepacia	-	1/1	1/1
P. mallei	-	2/2	2/2
P. putida	-	1/1	1/1
Serratia liquefaciens	-	1/1	1/1
S. rubidaea	-	1/1	1/1
Total	122/150 (81.3%)	30/35 (85.7%)	152/185 (82.2%))

Lower Respiratory Infections

Forty-five investigators, 28 domestic and 17 foreign, entered a total of 144 patients, 95 domestic and 49 foreign, with lower respiratory tract infections into this multicenter uncontrolled clinical trial. The criteria for selection and exclusion of patients with lower respiratory tract infections in this study were similar to those used in the multicenter controlled study reviewed earlier. Sputum smears for quantitative PMN and epithelial cell counts were done only in a few patients. One hundred seventeen patients, 80 domestic and 37 foreign, were evaluable for efficacy. The demographic characteristics of these patients are presented in Table I. The majority of patients had pneumonia, and had underlying conditions, such as chronic obstructive pulmonary disease (COPD), chronic bronchitis, pulmonary fibrosis, or cystic fibrosis.

As noted in Tables II and III, Pseudomonas aeruginosa and Haemophilus influenzae were the gram-negative pathogens most frequently isolated prior to therapy. The susceptibility of clinical isolates was tested by the disc diffusion or tube dilution method. All gram-negative isolates in the evaluable patients were reported to be susceptible to the test drug. Six isolates (3 P. aeruginosa, 2 H. influenzae, and 1 Hafnia alveii) had intermediate susceptibility to aztreonam as shown by the disc-diffusion method. In foreign studies, some investigators designated pathogens as susceptible, but zone diameters or the MICs were not given.

The dosage regimens in the domestic and foreign studies were 1 to 2 g every 8 hours, and 1 to 2 g every 8 to 12 hours, respectively. The drug was administered intravenously to all except one in the domestic study population, and to 60% (23 of 37) in the foreign study population. The duration of therapy was from 5 to 14 days in most patients. The mean duration of therapy was 11 days. Two domestic patients received prolonged aztreonam therapy; one cystic fibrosis patient with pneumonia and the other with lung abscess; these were treated for periods of 97 and 47 days, respectively. Some patients received antimicrobial agents effective for obligate anaerobes and gram-positive pathogens.

Clinical and laboratory monitoring of patients in this study was similar to that in the controlled study. The duration of post-therapy follow-up, however, was shorter in this uncontrolled study.

As shown in Table III, the microbiologic eradication rates were 92% (22/24) for H. influenzae, 80% (12/15) for K. pneumoniae , 30% (14/46) for P. aeruginosa, 78% (7/9) for E. coli, 75% (6/8) for P. mirabilis, and 80% (8/10) for Enterobacter species, including E. cloacae and E. aerogenes. The results were similar to those seen in the multicenter controlled study of LRTI. The lowest eradication rate for Pseudomonas aeruginosa was partly due to persistence or re-emergence of this microorganism in the majority of cystic fibrosis (CF) patients. The emergence of aztreonam-resistant P. aeruginosa and Pseudomonas sp. during or after completion of therapy was documented in 6 patients, 4 domestic and 2 foreign, in whom microbiologic failure occurred. Although eradication of the pathogen were achieved in few CF patients, the clinical responses were more favorable. Reinfections occurred in 4 evaluable patients; the microorganisms were S. pneumoniae in 3, and H. influenzae in 1. The microorganisms causing superinfection (or colonization) were S. aureus in 5, Enterobacter species in 5, Acinetobacter sp. and P. aeruginosa in 2 cases each, and S. epidermidis in 1, as shown in the applicant's Table 30 (Vol. 3.3) Favorable clinical responses (clinical improvement and partial responses) were seen in 89% (104/117) of this study population.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious (Lower respiratory tract) Infections due to Gram-negative Organisms

Table I

	Domestic	Foreign	Total
Number of Investigators	28	17	45
Number of Patients Entered	95	49	144
Number of Patients Excluded frome Efficacy Evaluation	15	12	27
Reasons for Exclusion:			
Pretreatment bacteriologic criteria not met	4	9	13
Inappropriate follow-up	5	0	5
5 days of therapy	1	1	2
Concurrent use of antibiotics*	4	2	6
Infection site other than LRT	1	0	1
Number of Patients Evaluable for Efficacy	80	37	117
Demographic Characteristics:			
Sex			
Female	31	12	43
Male	49	25	74
Age			
Range	2 - 93	3 - 81	
Mean	58.6	56.5	
Race			
Black	11	0	11
Caucasian	68	37	105
Other	1	0	1
Clinical Diagnosis:			
Pneumonia	69	24	93
Bronchitis	9	12	21
Lung abscess	2	1	3
Underlying cardiopulmonary diseases	44	23	67
Dosage Regimen(IV or IM):	1-2 g q 8 h	1-2 g q 8 -12 h	
IV	77	22	99
IM	1	13	14
IV/IM	2	2	4
Duration of Therapy (days):			
Range	3 - 97	6 - 20	
Mean	11.6	11.2	
Concurrent use of Antibiotics (clindamycin or metronidazole)	16	2	
(other antibiotics for gram-positive microorganims)	10	4	

* Antibiotics effective for gram-negative microorganisms.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (LRTI) due to Aerobic Gram-negative Organisms

Table II
Lower Respiratory Tract Infection

Microbiologic Response

Pathogen	No. Eradicated*/No. of Patients Treated		
	Domestic	Foreign	Total
Single Pathogen:			
P. aeruginosa	11/29 (38%)	2/8	13/37 (35%)
Pseudomonas sp.	-	1/6	1/6
H. influenzae	12/12	8/10	20/22 (91%)
Haemophilus sp.	1/1	-	1/1
K. pneumoniae	4/5	4/4	8/9 (89%)
K. oxytoca	1/1	-	1/1
S. marcescens	4/5	-	4/5
Serratia sp.	-	1/1	1/1
E. coli	3/4	1/2	4/6
E. aerogenes	2/2	1/1	3/3
E. cloacae	1/1	-	1/1
E. hafniae (Hafnia alvei)	1/1	-	1/1
Enterobacter sp.	-	1/1	1/1
P. mirabilis	2/2	-	2/2
C. freundii	2/2	-	2/2
C. diversus	1/1	-	1/1
Multiple Pathogens:			
E. coli + P. stuartii	1/1	-	1/1
E. coli + P. aeruginosa*	-	0/1	0/1
E. coli + K. pneumoniae	-	1/1	1/1
E. aerogenes + E. cloacae	1/1	-	1/1
E. aerogenes + P. mirabilis	1/1	-	1/1
E. cloacae + K. pneumoniae	0/1	-	0/1
E. cloacae + P. aeruginosa*	0/1	-	0/1
H. Influenzae + K. pneumoniae	1/1	-	1/1
H. influenzae + P. aeruginosa*	0/1	1/1	1/2
K. pneumoniae + C. diversus	1/1	-	1/1
K. pneumoniae + P. mirabilis	1/1	-	1/1
K. pneumoniae + P. aeruginosa	0/1	-	0/1
P. aeruginosa* + P. mirabilis	0/3	-	0/3
P. aeruginosa* + Serratia sp.	-	0/1	0/3
E. aerogenes* + P. mirabilis* + P. aeruginosa	0/1	-	0/1
Total (single + multiple)	51/80 (63.8%)	21/37 (56.8%)	72/117 (61.5%)

*Eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.

** The microorganisms not eradicated.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (LRTI) due to Aerobic Gram-negative Organisms

Table III

Lower Respiratory Tract Infections

Microbiologic Response

Pathogen	No. Isolates Eradicated/No. Treated		
	Domestic	Foreign	Total
P. aeruginosa	12/36 (33%)	2/10	14/46 (30%)
Pseudomonas sp.	-	1/6	1/6
H. influenzae	14/14	8/10	22/24 (92%)
Haemophilus sp.	1/1	-	1/1
K. pneumoniae	7/10	5/5	12/15 (80%)
K. oxytoca	1/1	-	1/1
S. marcescens	4/5		4/5
Serratia sp.	-	2/2	2/2
E. coli	4/5	3/4	7/9
E. aerogenes	4/5	1/1	5/6
E. cloacae	3/4	-	3/4
E. hafniae (Hafnia alvei)	1/1	-	1/1
Enterobacter sp.	-	1/1	1/1
P. mirabilis	6/8	-	6/8
C. freundii	2/2	-	2/2
C. diversus	2/2	-	2/2
Total	61/94 (64.9%)	23/39 (58.9%)	84/133 (63.2%)

*Eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (LRTI) due to Aerobic Gram-negative Organisms

Table III

Lower Respiratory Tract Infections

Microbiologic Response

Pathogen	No. Isolates Eradicated/No. Treated		
	Domestic	Foreign	Total
P. aeruginosa	12/36 (33%)	2/10	14/46 (30%)
Pseudomonas sp.	-	1/6	1/6
H. influenzae	14/14	8/10	22/24 (92%)
Haemophilus sp.	1/1	-	1/1
K. pneumoniae	7/10	5/5	12/15 (80%)
K. oxytoca	1/1	-	1/1
S. marcescens	4/5		4/5
Serratia sp.	-	2/2	2/2
E. coli	4/5	3/4	7/9
E. aerogenes	4/5	1/1	5/6
E. cloacae	3/4	-	3/4
E. hafniae (*Hafnia alvei*)	1/1	-	1/1
Enterobacter sp.	-	1/1	1/1
P. mirabilis	6/8	-	6/8
C. freundii	2/2	-	2/2
C. diversus	2/2	-	2/2
Total	61/94 (64.9%)	23/39 (58.9%)	84/133 (63.2%)

*Eradication was assumed where clinical improvement together with absence of sputum production was seen during and/or post-therapy.

Urinary Tract Infections (UTI)

In this open single-drug study, 55 investigators, 33 domestic and 22 foreign, treated a total of 171 patients, 128 domestic and 43 foreign, with UTIs caused by gram-negative pathogens. Data on 109 patients, 80 domestic and 29 foreign, were evaluable for efficacy. The demographic characteristics of those patients are shown in Table I. The majority of patients were elderly, and had complicated upper UTIs (pyelonephritis). E. coli, P. aeruginosa, and K. pneumoniae were the most frequently isolated pathogens. Fourteen patients, 12 domestic and 2 foreign, had polymicrobial infections. Other rare pathogens usually seen in patients with nosocomial infections were also seen in this study population. Susceptibility of clinical isolates was tested by the disk-diffusion or tube dilution method. All of the gram-negative isolates were susceptible to the test drug in the evaluable patients.

The dosage regimen and the duration of therapy were similar to those in the controlled UTI studies of multi-dose aztreonam therapy. The majority of patients received the drug intravenously, at the dose of 1 to 2 g, every 8 hours for 5 to 14 days.

As in the evaluation of the multicenter controlled study of UTI, this reviewer analysed microbiologic results at the 4 to 6 week post-therapy follow-ups. The bacteriologic responses seen in the domestic and foreign study population are presented in Tables II and III. The findings were similar to those observed in the multicenter controlled study. The eradication of urinary pathogens, particularly of P. aeruginosa in patients with complicated and/or recurrent UTI was achieved in only a small proportion of these patients, as shown in Tables III and IV. The overall eradication rates were 69% (31/45) for E. coli, 92% (12/13) for K. pneumoniae, and 26% (7/27) for P. aeruginosa. Reinfections occurred in 20 patients, 14 domestic and 6 foreign. The re-infection rates were 23% (15/66) in complicated UTI and 12% (5/41) in uncomplicated UTI. The micro-organisms were enterococci (S. faecalis) in 8, S. aureus in 3, E. coli and P. mirabilis in 2 cases each, and K. pneumoniae, M. morganii, S. marcescens, P. vulgaris and Candida albicans in one case each. The gram-negative bacilli were susceptible to aztreonam. The micro-organisms causing superinfections were S. aureus in 3, Pseudomonas species in 2 and S. epidermidis, P. stuartii, and enterococci in one case each.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious (Urinary taract) Infections due to Gram-negative Organisms

Table I

	Domestic	Foreign	Total
Number of Investigators	33	22	55
Number of Patients Entered	128	43	171
Number of Patients Excluded frome Efficacy Evaluation	48	14	62
Reasons for Exclusion:			
Pretreatment bacteriologic criteria were not met	3	0	3
Iuadequate follow-up (No 4-6 weeks follow-up)	45	14	49
Number of Patients Evaluable for Efficacy	80	29	109
Demographic Characteristics:			
Sex			
Female	57	14	71
Male	23	15	38
Age			
Range	16 - 97	18 - 85	
Mean	59.9	59.4	
Race			
Black	30	0	30
Caucasian	49	29	78
Other	1	0	1
Clinical Diagnosis:			
UTI (not specified)	11	9	20
Pyelonephritis	49	18	67
Cystitis	20	2	22
Complicated UTI	45	21	66
Uncomplicated UTI	35	6	41
Not specified	0	2	2
Dosage Regimen (IV or IM):	1-2 g q 8 h	1-2 g q 8 -12 h	
Duration of Therapy (days):			
Range	5 - 30	6 - 21	
Mean	10.2	10.9	

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (UTI) due to Aerobic Gram-negative Organisms

Table II
Urinary Tract Infection

Microbiologic Response

Pathogen	No. Eradicated*/No. of Patients Treated**		
	Domestic	Foreign	Total
Single Pathogen:			
P. aeruginosa	2/14	4/8	6/22 (27%)
E. coli	20/29	8/9	28/38 (74%)
K. pneumoniae	7/8	1/1	8/9
Klebsiella sp.	-	1/1	1/1
S. marcescens	1/3	-	1/3
Serratia sp.	-	1/1	1/1
P. mirabilis	0/1	1/1	1/2
E. cloacae	0/1	0/1	0/2
C. diversus	1/1	-	1/1
Multiple Pathogens:			
E. coli + P. stuartii	0/1	-	0/1
E. coli + Providencia sp.	0/1	-	0/1
E. coli + Pseudomonas sp.	-	0/1	0/1
E. coli + K. pneumonisae	1/1	-	1/1
E. coli + S. marcescens	1/1	-	1/1
Enterobacter sp.+ P. mirabilis***	-	0/1	0/1
E. cloacae + P. mirabilis	1/1	-	1/1
K. pneumoniae + K. oxytoca	1/1	-	1/1
K. pneumoniae + P. aeruginosa***	0/1	-	0/1
P. aeruginosa*** + P. stuartii	0/1	-	0/1
P. aeruginosa*** + P. rettgeri	0/1	-	0/1
P. aeruginosa*** + P. mirabilis	0/1	-	0/1
M. morganii + P. stuartii	1/1	-	1/1
E. coli + K. pneumoniae + P. aeruginosa	1/1	-	1/1
Total (single + multiple)	37/69 (54%)	16/24 (67%)	53/93 (57%)

*At 4-6 weeks after completion of therapy.
** Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.
*** The microorganism not eradicated.

Protocol 18554-16: Evaluation of Aztreonam in the Treatment of Serious Infections (UTI) due to Aerobic Gram-negative Organisms

Table III

Urinary Tract Infection

Microbiologic Response*

Pathogen	No. of Isolates Eradicated/No. of Patients Treated		
	Domestic	Foreign	Total
E. coli	23/35 (66%)	8/10 (80%)	31/45 (69%)
K. pneumoniae	11/12 (92%)	1/1	12/13 (92%)
K. oxytoca	1/1	-	1/1
Klebsiella sp.	-	1/1	1/1
P. aeruginosa	3/19	4/8	7/27 (26%)
Pseudomonas sp.	-	0/1	0/1
P. mirabilis	2/3	1/2	3/5
S. marcescens	2/4	..	2/4
Serratia sp.	-	1/1	1/1
E. cloacae	1/2	0/1	1/3
Enterobacter sp.	-	1/1	1/1
Providencia stuartii	1/2	-	1/2
P. rettgeri	1/1	-	1/1
Providencia sp	0/1	-	0/1
Citrobacter diversus	1/1	-	1/1
Morganella morganii	1/1	-	1/1
Total	47/82 (57%)	17/26 (65%)	64/108 (59%)

* At 4-6 week after completion of therapy.

Table IV

Bacteriologic Cure at 4-6 Weeks Post-Therapy

UTI	Number Cured /Number Treated*		
	Domestic Study	Foreign Study	Total
complicated UTI	14/37 (38%)	11/18 (61%)	25/55 (45%)
Uncomplicated UTI	23/33 (70%)	5/6 (83%)	28/39 (72%)

* Patients who developed superinfection during therapy or reinfection within 5-9 days post-therapy but had no further follow-up were not included.

The safety of aztreonam was assessed in patients with the infections of lower respiratory tract, urinary tract, skin and skin structures, and septicemia. The adverse reactions encountered in patients with infections of the bone and joints, intra-abdominal and gynecologic organs were reviewed earlier in the appropriate sections. The adverse reactions which were considerered as possibly or probably related to drug therapy were as follows:

Adverse Reactions	LRT	UTI	Septicemia	Skin/ Skin Structures
Clinical:				
Dermatologic:				
Exfoliative dermatitis	0	0	0	1
Rash	2	8	4	3
Purpura	1	0	0	0
Pruritus	0	1	1	0
Gastrointestinal:				
Nausea/vomiting	2	3	0	2
Diarrhea	0	6	1***	3
Taste alteration	0	2	0	1
Abdominal cramps	0	1	0	0
Hepatic:				
Jaundice	0	0	0	1
Local:				
Thrombophlebitis/ phlebitis	3	8	5	5
Other (pain, swelling, or warm feeling at injection sites)	0	5	0	2
CNS:				
Dizziness	0	1	0	1
Headache	0	1	0	0
seizure	1	0	0	0
Miscellaneous:				
Oral lesion (ulceration)	0	1	0	0
Vaginitis (Candida)	0	0	0	1
No. of Patients*	13	36	10	19
Laboratory**				
Elevated transaminases (ALT/AST)	8	6	5	4
Elevated alkaline phosphatase	0	1	0	0
Eosinophilia	6	4	2	2
Thrombocytopenia	0	0	0	1
Prolonged PT/PTT	1	0	1	1
Elevated serum creatinine	1	0	0	0
Elevated LDH	0	0	1	0
No. of Patients	15	11	9	8

*Some patients had more than one adverse effect.
** Patients with laboratory abnormalities prior to therapy were not included.
*** Caused by Clostridium difficile.

In twenty-two patients aztreonam was discontinued by the investigators because of adverse reactions.

Deaths:

A total of 73 deaths, 53 domestic and 20 foreign, occurred in seriously ill patients who received aztreonam for the treatment of the infections of the lower respiratory tract, urinary tract, skin/skin structures and septicemia, during therapy and post-therapy follow-up. The findings in this study indicated that the mortality rate remains high in elderly hospitalized patients with life-threatening gram-negative infections, inspite of potent antimicrobials currently available. This reviewer concurs with the applicant's conclusions that the deaths occurring during or after aztreonam therapy appear to be either to the severity of the infectious disease process, the concurrent lesions, and/or the age of the patients, and that there appeared to be no instance where death were directly attributable to the use of this new drug. It, however, should be noted that deaths occurred in a few patients due to fatal infections caused by aztreonam-resistant gram-positive pathogens.

Overall Conclusions and Recommendations:

Results of the multicenter, randomized controlled studies (domestic and foreign) of aztreonam vs. approved antibiotics (spectinomycin, cefamandole, or aminoglycosides) support the conclusions that the efficacy and safety of this new monobactam antibiotic is comparable to or superior to that of the control drugs used in the treatment of the following aerobic gram-negative infections:

1. Uncomplicated gonococcal infections of urethra and cervix.

2. Urinary tract infections (complicated and uncomplicated) caused by E. coli, P. mirabilis, Klebsiella pneumoniae, E. cloacae, Pseudomonas aeruginosa, Serratia marcescens, K. oxytoca*, Citrobacter species*, and indole-positive Proteus species.

   Note: The indication, a single-dose intramuscular aztreonam therapy for uncomplicated lower urinary tract infection (cystitis), is not approvable, since results of a multicenter, randomozed comparative study indicated that the single-dose aztreonam therapy appeared to be less effective than the conventional multi-dose amoxicillin therapy.

3. Lower Respiratory tract infections (LRTI) caused by E. coli, Haemophilus influenzae, K. pneumoniae, P. mirabilis, Enterobacter species, Serratia marcescens, and P. aeruginosa.

Results of the multicenter uncontrolled studies, domestic and foreign, were similar to those found in the controlled, randomized studies of urinarytract and lower respiratory infections.

In the treatment of intra-abdominal infections caused by aerobic gram-negative microorganisms, results of a multicenter, randomized, controlled study in a limited number of patients suggested superior efficacy of this drug over the control drug, an aminoglycoside. In this study, antianaerobic agents were administered concomitantly and appropriate surgical interventions were made in the study population. Results of an uncontrolled study were supportive of the findings of the limited controlled study. However, more data are needed to confirm the results seen in these studies.

In the treatment of septicemia and skin/skin structure infections caused by susceptible strains of gram-negative aerobic microorganisms, results of uncontrolled (non-comparative), multicenter studies suggested that aztreonam is effective and safe. However, controlled clinical trials of this new monobactam antibiotic vs. currently approved antibiotics are desirable to ascertain the usefulness of this new antibiotic and to confirm the results seen in the uncontrolled studies.

The approval of the indications, septicemia, and skin/skin structure infections can only be considered with the inclusion of the following statement: "Although data from controlled clinical trials are not available, aztreonam has been shown in uncontrolled studies to be effective in the treatment of the following indications:

Septicemia caused by Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Proteus mirabilis*, Serratia marcescens*, Enterobacter species .

Skin and Skin structure infections caused by E. coli, P. mirabilis, S. marcescens, Enterobacter species, Pseudomonas aeruginosa, and K. pneumoniae.

The number of patients with bone and joint infections (osteomyelitis and septic arthritis) treated in the uncontrolled studies was small, and furthermore the duration of follow-up was less than optimum. Controlled clinical trials are also needed for the approval of this indication.

The number of patients with gynecologic infections studied in this open study was also small, as in the controlled study. Further controlled studies of aztreonam in which adequate post-therapy follow-up has been made are needed to support the favorable findings seen in the very limited numbers of patients studied so far.

At present, the indications: intra-abdominal infections, obstetric and gynecologic infections, and bone and joint infections are not approvable, since the clinical data submitted are inadequate to support the claims.

Adverse reactions observed were usually mild and transient, although in a few patients, the drug was discontinued by investigators because of adverse reactions. The most common adverse effects were local reaction at the site of infusion or injection, dermatologic (rash), gastrointestinal (nausea/vomiting or diarrhea), transient increases in transminases (ALT/AST) and eosinophilia. Superinfection or colonization with gram-positive organisms, particularly enterococci (S. faecalis) was encountered in patients with complicated UTI. The incidence, however, was similar to that with the control drug, a second generation cephalosporin. Superinfection or reinfection caused by the gram-positive organism, Staphylococcus aureus and S. pneumoniae occurred in a few aztreonam-treated patients with lower respiratory tract infections. Awareness of these effects is critical in the management of hospitalized patients, particularly geriatric or immunocompromised patients, receiving this drug. Deaths occurred in many seriously ill hospitalized patients who received the test and the control drugs. None of the deaths were directly attributable to drug therapy. The incidence of pulmonary embolism and deaths was similar in the aztreonam treatment group and the control-drug group.

F. Min

F. Min, M.D.

Orig Form 5 50-580
HFN-815, HFN-815/CSO
HFN-340, HFN-535
HFN-815/RNorton
HFN-815/FMin:js/12/5/85
4586b

ET 1/27/86 See my handwritten note below signature on Group Leader's Comments

DRD 26Dec85 see Group Leader's Comments dated 26Dec85

PHARM REVIEW

REVIEW & EVALUATION OF PHARMACOLOGY & TOXICOLOGY DATA

NDA 50-580 (Amendment , dated 2/27/85)

Date Review Completed: 4/5/85

Applicant: E.R. Squibb & Sons, Inc., New Brunswick, NJ

Drug: Aztreonam for injection (Azactam^R)

Category: Synthetic antibiotic

Additional Preclinical Studies

General Pharmacology of Aztreonam (AZT)

Lab Perf. Study: NRI, Life Sciences, Japan

The following results have been reported:

1. AZT prolonged hexobarbital-induced sleep time at 750mg/kg (IV) in mice (ONEL* = 270mg/kg) and increased the amplitude of EEGs at 100mg/kg (IV) in rabbits (ONEL = 37mg/kg). The drug did not affect motor activity, the rotatory rod test, acetic acid writhing, convulsions, body temp. or spinal reflex.

*ONEL = Observed No Effect Level

2. AZT (100mg/kg & above, IV) increased peripheral blood flow, respiratory frequency & HR, and decreased BP. A metabolite of AZT, AQ 26,992 (100mg/kg or above, IV) also produced similar effects. Arginine alone at high doses (585 mg/kg, IV) also produced similar effects. AZT had no effect on isolated guinea pig atria.

3. AZT did not affect the BP response to vasoactive amines in anesthetized dogs. Given IV, the drug (750mg/kg) or arginine alone (585mg/kg) caused a slight suppression of the nictitating membrane contractility in cats.

4. AZT (10^{-4}g/ml) slightly inhibited the motility of isolated ilea. Drug-induced contraction of the ileum, Ca^{2+}-induced contraction of the cecum, and drug-induced contraction of the trachea and vas deferens were also not affected by AZT at 10^{-4}g/ml conc'n. Oxytocin-induced contractions of pregnant uteri and motility of pregnant or non-pregnant uteri were not altered by AZT (10^{-4}g/ml conc'n. Oxytocin-induced contractions of uteri isolated during the diastrous period were slightly inhibited.

5. AZT (750mg/kg, IV) transiently inhibited the in situ uterine motility of non-pregnant rats & oxytocin-induced contractions in pregnant or non-pregnant rats. At 100mg/kg, these effects were not seen.

The in situ motility of the stomach was inhibited immediately after admin. of AZT (100 mg/kg & above), but recovered in about 30 min. in most cases. At 750mg/kg, IV, it accelerated the transport through the digestive tract in mice. Cephazoline (750mg/kg) increased gastric motility, but had no effect on intestinal propulsion.

6. AZT (100-750mg/kg, IV) had no sig. effects on gastric secretion, neuromuscular transmission, anti-inflammation, pancreatic secretion, platelet aggregation, blood coagulation or fibrinolytic parameters.

7. AZT (750mg/kg, IV) increased urine volume & K^+ excretion rate and decreased RPF in anesthetized dogs. Arginine alone (585mg/kg, IV) also had similar effects. Cephazoline (750mg/kg) decreased RPF & increased FF.

8. AZT at high dose (750mg/kg, IV) slightly increased the serum GOT & GPT levels (1/3 animals). CLE & glutathione levels were unaffected. "No effect" level was reported to be 270mg/kg. Arginine alone (585mg/kg, IV) also showed slight increases in GOT & GPT levels. Both cephazoline & AZT increased the ICG retention rate at 750mg/kg dose level.

9. AZT (37-750mg/kg, IV) increased bile secretion in rats.

Comments:

These Japanese studies were performed to satisfy requirements for registration of the drug in that country. Results mostly similar to these have been reported in the original NDA application and have been reviewed before approval of the application. No significant adverse effects not reported earlier could be found in these results. No action is necessary.

Syed N. Alam

cc: ~~Orig. NDA~~
HFN-815
HFN-815/MO
CSO
HFN-340
HFN-815/SNAlam/smc/4/26/85
R/d init.by:JMDavitt
3844b

REVIEW & EVALUATION OF PHARMACOLOGY & TOXICOLOGY DATA

NDA 50-580 (Amendment, dated 12/28/84)

Applicant: .R. Squibb & Sons, Inc., New Brunswick, NJ

Drug: Azactam^{R} (aztreonam for injection)

Category: Synthetic antibiotic

Preclinical Studies

Lab Performing Studies: The sponsor

1. Polyacrylamide Gel Electrophoresis of Human Serum Proteins after Incubation with ^{14}C-Aztreonam & ^{14}C-Penicillin G:

   ^{14}C-Aztreonam (SQ 26,776) & ^{14}C-penicillin G, each at a conc'n equimolar to 100 ug/ml of ^{14}C-aztreonam, were separately incubated with human sera obtained from volunteers (3) at 37°C for 24 hrs, then subjected for gel electrophoresis & fluorography.

   Results showed that most of the radioactivity (both aztreonam & penicillin G) that was covalently bound to proteins was associated with the albumin fraction. A much smaller amount was bound to alpha-globulin fraction.

2. Acute IP Toxicity Study in Mice:

   Species & No. of Animals: Male albino mice; 4 groups, 10/sex/group

   Route of Admin.: IP

   Methods: Acute LD_{50} values of aztreonam, one of its isomers and some derivatives were determined by IP injections of suspension of the drugs to CD-1 male albino mice. The results are shown in the table below.

Group	Material Tested	LD_{50}, mg/kg (95% CI)
1	Aztreonam	1380 (1120-1700)
2	Aztreonam E isomer (SQ 28,429)	730 (520-1020)
3	Aztreonam ethyl ester (SQ 27,412)	2000 (1450-2760)
4	Desulfonated SQ 26,992 (SQ 29,294)	1740 (1225-2470)

   Period of Observation: 14 days

   Toxicity Symptoms: Toxic signs included decreased motor activity, decreased respiratory rate, convulsions, loss of strength in hindquarters, terminal collapse & weight loss.

Evaluation:

In a previous submission (6/1/83; IND 18,554), the sponsor reeported the covalent binding of aztreonam to human serum proteins. This has been confirmed in the present submission. Additionally, binding has been shown to occur predominantly with albumin fractions and very little with the alpha-globulin fractions. Penicillin G has also been shown to bind covalently with the serum albumin fractions. Thus, the covalent binding to proteins is not unique to aztreonam.

The IP LD_{50} determination in mice indicated that the E isomer of aztreonam (SQ 28,429) was twice as toxic as aztreonam and the ethyl ester (SQ 27,412) and desulfonated SQ 26,992 (SQ 29,294) were equitoxic to or slightly less toxic than aztreonam.

No action is necessary.

final

S.N. Alam, Ph.D.

cc: Orig. NDA
HFN-815 2T 4/29/85
HFN-815/MO
CSO
HFN-340
HFN-815/SNAlam/smc/4/23/85
R/d init.by:JMDavitt
3798b

REVIEW & EVALUATION OF PHARMACOLOGY & TOXICOLOGY DATA

NDA 50-580 (Amendment, dated 10/3/84)

Date Review Completed: 10/29/84

Applicant: E.R. Squibb & Sons, Inc., New Brunswick, NJ

Drug: Aztreonam (Azactam^{R})

Category: Synthetic antibiotic

Preclinical Studies:

1. 4-Week IV (Infusion) Toxicity in Dogs

   Lab Performing Study: Dr. F. Leuscher, Laboratory of Pharmacology & Toxicology, Hamburg, W. Germany.

   Material Tested: Aztreonam (2%) in 0.9% NaCl sol'n

   Species & # Animals: Beagle dog (male); 2/group

   Dose Levels: 0 (control), 10, 25 or 62.5ml/kg equiv. to 0, 200 (LD), 500 (MD) or 1250 (HD) mg/kg

   Route & Duration: IV infusion over a 3-hr period, once/day for 28 consecutive days

   Results

   Clinical Observations: Occasional pultaceous feces in all animals and vomiting in 1 HD dog (only once after the first infusion) were observed.

   Mortality: None

   Body Wt; Food & Water Intake: Normal

   Hematology: Increased neutrophils (group mean) and decreased lymphocytes were seen in the MD & HD gps. Platelet counts were low in all drug-treated groups. In 1 HD dog, there was more than 50% reduction in platelet counts compared to pre-test value. ESR increased in a dose-related manner in the MD & HD gps.

   Clinical Chemistry: All parameters measured were within normal range.

   Urinalysis: No unusual findings

   EKG; BP: No abnormal findings

   Ophthalmoscopy & Hearing Test: No ocular changes or impairment of auditory acuity was reported.

Gross Pathology: Unremarkable except for small areas of bleeding at the infusion sites in all groups (including controls).

Organ Wt: Abs. spleen wts in the LD & HD groups were lower than in controls. All other organs weighed were within normal variations.

Histopathology: Except for hemorrhage at the injection sites in all groups, no pathological findings have been reported.

2. IV Perinatal & Postnatal Study in Rats

Lab Performing Study: NRI Life Science, Kanagawa, Japan

Materials Tested: SQ 26,776 with arginine (10:8) in dist. water

Species & # Animals: SD rats; 20/group

Dose Levels: 0 (control), 100 (LD), 270 (MD) or 750 (HD) mg/kg

Route, Frequency & Duration: IV injection; once/day from day 17 of gestation to day 20 of lactation (day of copulation was assigned as day 0 of gestation).

Results

Effects on Dams

a) During Gestation: No death or abnormalities were noted. Body wts were similar in all gps. Food intake decreased significantly in the treated animals between day 17 & 20 of gestation.

b) At Delivery & During Lactation: One HD pregnant animal died on day 23 of gestation without delivering. Grossly, adrenal hypertrophy was noted in this animal. Microscopic findings included vacuolation of both hepatocytes & tubular epithelium with hyaline droplets. Hypertrophy of the adrenal cortex and recession of the thymic & splenic white pulps were also reported. No abnormalities were reported in other dams during delivery & lactation. The MD & HD animals showed sig. increase in body wt (compared to controls) during lactation.

c) Autopsies on Day 21 of Lactation: Dilatation of the renal pelvis & mastitis (1 LD), SC edema on all limbs (1 HD), increased abs. & rel. cecum wt in all 3 drug-treated gps compared to controls and significantly increased abs. & rel. liver wts (MD & HD) were reported.

d) Reproduction Data (F_0 Dams) & Observations on F_1 Rats: These are shown in the table below.

Parameters	Control	Low Dose	Mid Dose	High Dose
# implantations (mean)	14.8	14.4	14.9	15.1
Mean length of gestation (days)	21.8	21.8	21.8	21.8
Mean # F_1 alive - Day 0	13.8	13.3	14.0	14.2
Day 4	13.4	12.6	13.8	14.1
Day 21	9.6	9.1	9.7	9.5
Mean # F_1 dead (%) - At birth	1.0	3.1	1.3	0.0
Day 1	0.4	0.0	0.4	0.4
Days 2-4	2.2	4.9	1.8	0.7
Day 21	0.0	0.0	3.0	2.7
Male/female ratio	1.13	0.97	1.01	0.93
Live birth index (%)	93.2	92.0	94.3	94.1
Viability index	97.5	95.1	97.9	98.9
Weaning index	100.0	100.0	97.0	97.3
# F_1 with malformation	0	0	0	0

No significant treatment effects were seen in F_1 animals at 21-day necropsy, when organ wts, functional development, behavioral developments, emotional activity, learning ability & reproductive performance were examined. An increase in cecum wt in almost all F_1 neonates were seen in the treatment gps.

Effects on F_1 Dams & F_2 Fetuses: (Day 20 necropsy)

These are shown in the Table 31 (attached).

Effects on F_2 Dams & Their Pups:

These are shown on Table 32 (attached).

One F_2 LD rat had anophthalmia; one F_2 HD rat had renal pelvis dilatation. No other abnormalities were reported in F_2 fetuses or pups.

Comments: Vacuolation of hepatocytes & tubular epithelium among the drug-treated animals reported here have been found previously in subacute animal toxicity studies. Cecum enlargement has also been reported before. No other unusual adverse findings have been reported in either the 4-week IV toxicity study or the reproduction study performed in rats. Thus, the "approvable" status of the NDA remains unchanged.

There is a discrepancy in Table 32 (reproduction study). On p. 25, the report says that "Tables 32, 33, 34 & 35 show various findings relating to F_2 rats." However, in Table 32 the data refer to F_1 rats. The sponsor should be asked to clear up this confusion.

cc: Orig. NDA
HFN-815 - 11/21/84
HFN-815/MO
CSO
HFN-220
HFN-815/SNAlam/smc/11/8/84
R/d init.by:JMDavitt
Attachments (2)
2176b

Syed N. Alam, Ph.D.

TABLE- 31 OBSERVATIONS OF FETUSES FROM F1 DAMS IN PERINATAL AND POSTNATAL STUDY OF S026,776

COMPOUND	CONTROL	S026,776		
DOSE (MG/KG)		100	270	750
NO. OF LITTERS (F1)	11	9	10	11
NO.OF CORPORA LUTEA TOTAL	(179)	(140)	(158)	(171)
MEAN ± S.D.	16.3± 2.2	15.6± 2.1	15.8± 2.3	15.5± 3.0
PREIMPLANTATION LOSS (%)	18/179 (10.1)	9/140 (6.4)	14/158 (8.9)	27/171 (15.8)
NO.OF IMPLANTATIONS TOTAL	(161)	(131)	(144)	(144)
MEAN ± S.D.	14.6± 1.6	14.6± 2.7	14.4± 2.1	13.1± 2.6
NO.OF DEAD IMPLANTATIONS				
RESORPTIONS (%)	8/161 (5.0)	2/131 (1.5)	2/144 (1.4)	2/144 (1.4)
PLACENTAL REMNANTS (%)	0/161 (0.0)	0/131 (0.0)	0/144 (0.0)	2/144 (1.4)
MACERATED FETUSES (%)	0/161 (0.0)	0/131 (0.0)	0/144 (0.0)	0/144 (0.0)
DEAD FETUSES (%)	0/161 (0.0)	0/131 (0.0)	0/144 (0.0)	0/144 (0.0)
TOTAL DEAD IMPLANTATIONS (%)	8/161 (5.0)	2/131 (1.5)	2/144 (1.4)	4/144 (2.8)
NO.OF LIVE FETUSES TOTAL	(153)	(129)	(142)	(140)
MEAN ± S.D.	13.9± 1.9	14.3± 2.4	14.2± 2.0	12.7± 2.8
SEX RATIO (MALE / FEMALE)	71/ 82 (0.87)	59/ 70 (0.84)	65/ 77 (0.84)	76/ 64 (1.19)
BODY LENGTH (MM) MALE MEAN ± S.D.	38.3± 0.9	38.2± 0.7	38.9± 0.7	38.9± 0.9
FEMALE MEAN ± S.D.	37.6± 1.0	37.5± 0.6	38.1± 0.6	38.2± 1.2
TAIL LENGTH (MM) MALE MEAN ± S.D.	13.7± 0.5	13.2± 0.6	14.0± 0.6	13.6± 0.7
FEMALE MEAN ± S.D.	13.6± 0.5	13.1± 0.6	13.8± 0.5	13.5± 0.8
BODY WEIGHT (G) MALE MEAN ± S.D.	3.69± 0.25	3.67± 0.14	3.77± 0.19	3.85± 0.31
FEMALE MEAN ± S.D.	3.49± 0.28	3.52± 0.11	3.51± 0.17	3.60± 0.34
PLACENTAL WEIGHT (MG) MALE MEAN ± S.D.	498.± 41.	478.± 74.	475.± 64.	511.± 58.
FEMALE MEAN ± S.D.	472.± 36.	461.± 75.	453.± 54.	499.± 70.
ADHESION OF PLACENTA	0	1	0	0
NO.OF FETUSES WITH MALFORMATIONS	0	0	0	0

* P<0.05, ** P<0.01 SIGNIFICANT DIFFERENCE FROM CONTROL (STUDENT'S T-TEST)
* P<0.05, ** P<0.01 SIGNIFICANT DIFFERENCE FROM CONTROL (ASPIN-WELCH'S T-TEST)
* P<0.05, ** P<0.01 SIGNIFICANT DIFFERENCE FROM CONTROL (WILCOXON'S RANK SUM TEST)

Table 32

COMPOUND	CONTROL	SQ26,776		
DOSE (MG/KG)		100	270	750
NO. OF LITTERS (F1)	7	7	6	7
IMPLANTATIONS TOTAL	(105)	(110)	(90)	(98)
MEAN ± S.D.	15.0± 0.8	15.7± 1.1	15.0± 2.6	14.0± 2.2
MEAN LENGTH OF GESTATION PERIOD (DAY) MEAN ± S.D.	21.7± 0.5	21.9± 0.4	22.0± 0.0	21.9± 0.4
LIVE ALIVE F1				
POSTPARTUM DAY 0	(93)	(99)	(79)	(93)
MEAN ± S.D.	13.3± 2.4	14.1± 1.6	13.2± 2.6	13.3± 2.1
POSTPARTUM DAY 1	(93)	(99)	(70)	(93)
MEAN ± S.D.	13.3± 2.4	14.1± 1.6	11.7± 4.2	13.3± 2.1
POSTPARTUM DAY 4	(92)	(99)	(69)	(92)
MEAN ± S.D.	13.1± 2.2	14.1± 1.6	11.5± 4.1	13.1± 2.1
AFTER SELECTION	(70)	(70)	(53)	(70)
MEAN ± S.D.	10.0± 0.0	10.0± 0.0	8.8± 2.0	10.0± 0.0
POSTPARTUM DAY 21	(70)	(69)	(49)	(65)
MEAN ± S.D.	10.0± 0.0	9.9± 0.4	8.2± 1.8	9.3± 0.8 *
LIVE DEAD F1				
AT BIRTH	1/105	0/110	3/ 90	2/ 98
(%)	(1.0)	(0.0)	(3.3)	(2.0)
POSTPARTUM DAY 1	0/ 93	0/ 99	9/ 79	0/ 93
(%)	(0.0)	(0.0)	(11.4)	(0.0)
POSTPARTUM DAY 2-4	1/ 93	0/ 99	1/ 70	1/ 93
(%)	(1.1)	(0.0)	(1.4)	(1.1)
POSTPARTUM DAY 5-21	0/ 70	1/ 70	4/ 53	5/ 70
(%)	(0.0)	(1.4)	(7.5)	(7.1)
SEX RATIO OF SURVIVORS (MALE / FEMALE)	50/ 43 (1.16)	49/ 50 (0.98)	34/ 45 (0.76)	51/ 42 (1.21)
LIVE BIRTH INDEX (A)	93/105	99/110	79/ 90	93/ 98
(%)	(88.6)	(90.0)	(87.8)	(94.9)
VIABILITY INDEX (B)	92/ 93	99/ 99	69/ 79	92/ 93
(%)	(98.9)	(100.0)	(87.3)	(98.9)
WEANING INDEX (C)	70/ 70	69/ 70	49/ 53	65/ 70
(%)	(100.0)	(98.6)	(92.5)	(92.9)
GROSS F1 MALFORMATION	0	0	1 (AN.)	0

: P<0.05, :: P<0.01 SIGNIFICANT DIF FROM CONTROL (STUDENT'S T-TEST)
* P<0.05, ** P<0.01 SIGNIFICANT DIFF FROM CONTROL (ASPIN-WELCH'S T-TEST)
# P<0.05, ## P<0.01 SIGNIFICANT DIFFERENCE FROM CONTROL (WILCOXON'S RANK SUM TEST)

*Anophthalmia

REVIEW & EVALUATION OF PHARMACOLOGY & TOXICOLOGY DATA

NDA 50-580 (Original Submission, dated 1/19/84)

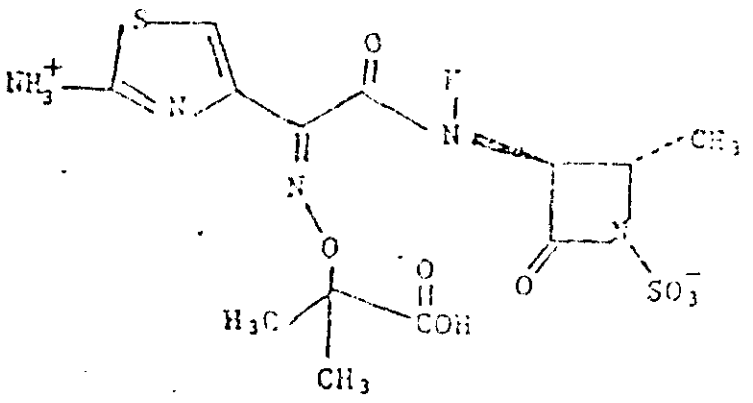
Date Review Completed: 6/21/84

Applicant: E.R. Squibb & Sons, New Brunswick, NJ

Drug: Proprietary Name: Azactam® (Azlactam®, Aztreonam)
Code Name: SQ 26,776

Chemical Name: (Z)-2-[[[(2-amino-4-thiazolyl)[[(2S,3S)-2-methyl-4-oxo-1-sulfo-3-azetidinly]carbamoyl]methylene]amino]oxy]-2-methylpropionic acid

Structure:



Category: Synthetic antibiotic

Composition:

*Amoun

Intended Route of Administration: Parenteral

Intended Use: Against infections primarily due to gram-negative bacteria

Related Submission: [redacted]

Preclinical Studies

The following studies contained in this NDA have been submitted in the past at various times in connection with applicant's [redacted] and amendments thereto, and have already been reviewed. These studies are listed in chronological order of submission. The number in ( ) after each study is the reference number used in the NDA for that study.

Original IND Submission

Pharmacol. Rev. dated 5/12/81:

1. In Vitro Activity of Aztreonam I. Antibacterial Activity (1)
2. In Vitro Activity of Aztreonam II. Characteristics and Factors Influencing Antibacterial Activity (2)
3. Stability Studies of Aztreonam (3)
4. Mode of Action of Aztreonam (6)
5. Evaluation of Aztreonum in Gram-Negative Systemic Infections in Mice (9)
6. A Pharmacokinetic Evaluation of Aztreonam (12)
7. Acute IV Cardiovascular and Renal Safety Study in Dogs (39)
8. Acute IP Toxicity in Rats, SQ 26,776 solution, 20% (35)
9. Acute IV Safety Test in Mice (25)
10. Acute IP Toxicity in Rats, SQ 26,776 Solution, 20% (35)
11. 30-Day Subcutaneous Toxicity Study in Rats (40)
12. 30-Day IV Study in Dogs (40)
13. Muscle Irritation Study in Rabbits (56)
14. Study of Pain in Dogs After IM Injection (57)
15. Compatability with Human Erythrocytes (55)
16. In Vitro Synergism and Antagonism Studies (2)

Amendments

Pharmacology Review dated 8/21/81:

2-Week SC Toxicity Study in Rats with Arginine Blend (41)

Pharmacology Review dated 10/31/81:

Acute Oral Toxicity of Arginine Blend in Mice

Pharmacology Review dated 2/26/82:

1. One-Month SC Toxicity Study in Rats (40)
2. One-Month IV Toxicity Study in Dogs (40)

3. Disposition of SQ 26,776-^{14}C after IM & IV Administration to Monkeys (15)

4. Acute Toxicologic Studies in Mice of the Interaction of SQ 26,775 with Ampicillin, Cefoxitin, Clindamycin, Gentamycin, Furosemide and Ethanol (27)

5. In Vitro Protein Binding of SQ 26,776-^{14}C to Rat, Dog, Monkey and Human Serum (22)

6. Covalent Binding of SQ 26,776-^{14}C and ^{14}C-Penicillin G to Human Serum Proteins (23)

Pharmacology Review dated 7/14/83:

1. In Vitro Testing of Immunological Cross-reactivity of Aztreonam with Other B-lactam Antibiotics

2. Acute Toxicologic Studies in Mice of the Interaction of SQ 26,776 with Probenecid, Tobramycin and Vancomycin (28)

3. Mutagenicity Evaluation of SQ 27,776 in the Mouse Lymphoma Forward Mutation Assay (52)

4. Disposition of SQ 26,776-^{14}C after IM & IV Administration to Rats (13)

5. Disposition of SQ 26,776-^{14}C after IV & SC Administration to Dogs (14)

6. SC 2-Generation Study of Reproduction & Development in Rats (47). Also see Pharm. Rev. dated 7/2/82.

Pharmacology Review dated 12/6/82:

1. Full Report on 2-Generation SC Reproduction Study (47)

2. A Segment II SC Teratology Study in Rats (48)

3. A Segment II SC Teratology Study in Rabbits (49)

4. A Segment III (Perinatal, Postnatal) Reproductior Study in Rats (50)

Pharmacology Review Dated 8/29/83:

Effect of Aztreonam/Arginine on Glomerular Filtration (Independent study of Dr. Brenner at Harvard Univ.)

Pharmacology Review dated 1/18/84:

Resubmission of Dr. Brenner's study with new interpretation

Preclinical Studies New in this NDA: (All studies performed by the applicant, unless otherwise indicated.)

Note: Throughout this review, SQ 26,776 will be abbreviated "SQ" and Aztreonam will be abbreviated "AZ".

Microbiology

See Microbiology Review. Since this is a Form 5 application which will be reviewed by the microbiologist, this section is not reviewed in its entirety. Only a few pertinent studies are summarized rather briefly.

1. In Vitro Activity of AZ,II: Characteristics & Factors Influencing Antibacterial Activity: AZ was shown to be very similar to other B-lactam antibiotics (cefmetazole, cefotaxime) in terms of environmental effects such as media, pH, inoculum size and presence of 50% human serum. The bactericidal activity of AZ was very similar to other B-lactam antibiotics in terms of both rate of killing and clearance. Resistance studies indicated that under appropriate selective pressure resistance develops. Studies in E. coli K 12 indicated that decreased membrane permeability might account for this resistance. AZ was compatible with several antibiotics of different spectra of activity (nafcillin, cloxacillin, erythromycin, vancomycin) and combinations of AZ with aminoglycosides was often synergistic.

2. Interaction of AZ & Other New B-Lactam Antibiotics with B-Lactamases from Gram-Negative Bacteria: AZ was shown to exhibit a high degree of stability to both plasmid- mediated and chromosomally-mediated B-lactamases from gram-negative bacteria. Stability determinations using B-lactamases from a variety of clinical isolates also indicated that AZ displayed good stability. Appreciable hydrolysis was observed only with the broad spectrum K 1 B-lactamase from Klebsiella pneumoniae and to a lesser extent, with the PSE-2 B-lactamase. AZ exhibited poor affinity for penicillinases and broad spectrum B-lactamases, but bound tightly to cephalosporinases. Studies with P-99 B-lactamases showed that AZ and moxalactam were potent inhibitors followed by cefotaxime and then by ceftazidime & cefoperazone.

3. Induction of B-Lactamase by AZ & Other B-Lactam Antibiotics: The induction studies were carried out with a wide selection of clinically important gram-negative organisms including highly inducible strains of Enterobacter, Proteus, Pseudomonas & Serratia. Maximum induction of -lactamases was caused by ampicillin & cefoxitin.

   Ceftazidime & cefotaxime also induced high levels of B-lactamase. AZ & cefaperazone were poor inducers in most organisms. AZ caused sig. induction in only two (both Proteus vulgaris) of the 46 isolates studied.

4. Evaluation of AZ in Model Infections:

   a) Urinary Tract Infection (Acute Pyelonephritis) in Mice: Acute pyelonephritis was induced in mice by injecting E. coli suspensions

(ampicillin sensitive & resistant) into the bladders of diuresed mice. AZ was found as effective as cefotaxime (ED_{50}s = $\leq$ 1.6mg/kg after 4 consecutive days of treatment) in treating these kidney infections.

b) Neutropenic Mouse Model: Mice made neutropenic by cyclophosphamide treatment were effectively protected by AZ against several gram-negative bacterial infections. For example, AZ had an ED_{50} of 5.9mg/kg against E. coli SC 12,677 and an ED_{50} of 1.4mg/kg in infections caused by B-lactamase positive E. coli SC 12,199. Against Pseudomonas infection, AZ was effective with ED_{50} of 56mg/kg.

c) Mixed Bacterial Infection: A 1:4 part AZ/nafcillin mixture was tested against a single gram-positive or gram-negative bacterial infection, as well as against mixed infections in normal and neutropenic mice. There was no antagonistic activity between these 2 antibiotics and the mixed infection (Staphylococcus-Serratia) was effectively treated with the combination.

d) Lower Respiratory Tract Infection in Rats: AZ was more effective than piperacillin & moxalactam in treating rat lung infections caused by Serratia marcescens (ED_{50} = 24.2mg/kg/day) and Pseudomonas aeruginosa (ED_{50} = 19.8mg/kg/day). Gentamicin had ED_{50} of 7.4-7.8 in this model.

e) Bacterial Meningitis in the Infant Rat: AZ was reported to be effective (ED_{50} = 5.6mg/kg/day) in treating an H. influenzae meningitis in infant rats. Cefoperazone, cefotaxime, ceftazidime, ceftizoxime & moxalactam were also effective with CD_{50}'s in the range of 3-8mg/kg/day. Ampicillin, piperacillin & chloramphenicol were essentially inactive in this model.

f) Surgical Wound Infection in Mice: AZ administered parenterally or topically in a cream base was effective against Proteus, Serratia & Pseudomonas infected surgical wounds in mice. Cefotaxime was not effective in this model, while gentamicin was very effective.

5. Evaluation of AZ in Hamster Colitis Model:

Method: Male, golden Syrian hamsters were injected IP with a single dose (100mg/kg) of the test compound. At selected times, filtrates of cecal contents were tested for cytotoxicity on MRC-5 fibroblasts for determination of Clostridium difficile toxin. The hamster model for colitis was established using clindamycin as reported by Chang et al. (Infect. Immun. 20: 526-529, 1978).

Results: AZ, in contrast to antibiotics like clindamycin, moxalactam & cefoperazone, did not induce antibiotic-associated colitis in hamsters. No C. difficile was recovered from the AZ-treated animals, although aerobic gran-negative rods were eliminated from the cecum by AZ treatment.

Pharmacology

1. Isolation and Identification of the Major Metabolite of AZ in Monkey Urine

Method: Six F cynomolgus monkeys were dosed IV or IM at 25mg/kg ^{14}C-labeled SQ and 0-4 hr urine was collected. The samples were combined, centrifuged, concentrated by evaporation & chromatographed by HPLC.

Results: HPLC showed 3 major peaks, one of which corresponded to unchanged SQ. The material in another peak was found to be unstable and was not analyzed further. The chromatographic fractions of the 3rd peak were combined & rechromatographed after evaporating to dryness. Again a single peak [both UV absorption & radioactivity (RA)] was obtained. The material was identified as SQ 26,992, a metabolite produced from the parent compound by opening of the β-lactam ring. This metabolite was identified by HPLC, NMR & Mass Spectrometry. It represented about 14% of the total urinary RA.

2. Quantitation of the ^{14}C-AZ Metabolites in Monkey Urine

Methods: Urine samples (0-24 hr) were obtained from 3 cynomolgus monkeys that were given single IM or IV 25mg/kg doses of ^{14}C-AZ with a washout period of 7 days between doses. The metabolites in the urine were analyzed by HPLC method. The isolated metabolites were than assayed by microbiological methods for antibacterial activity.

Results: Nearly identical chromatograms were obtained from urine of the monkeys after either route of admin. There was a total of 5 peaks (I-V), one of which was AZ. The relative distribution of these peaks (RA) is shown in the table below.

Compound	% Relative Distribution in Urine	
	IM	IV
Aztreonam (V)	73.6	78.2
SQ 26,992 (IV)	13.7	11.8
Metabolite III	2.9	2.7
Metabolite II	3.6	2.5
Metabolite I	1.2	0.8
Total	97.0	97.8

Bioassay using E. coli s.c. 12,155 showed that none of the metabolites had any antibacterial activity.

3. Tissue Distribution of ^{14}C-AZ after Single IM Administration to Rats

Methods: Gps of 6 rats (3/sex) were sacrificed at 0.25, 2, 6 & 24 hrs after IM admin. of single 50mg/kg doses of ^{14}C-AZ. Conc'ns of total RA were determined in serum & 24 tissues. Conc'ns of unchanged AZ were determined in serum, kidney, liver, lung, and in the contents of small & large intestines. Additionally, whole body autoradiography was carried out with some of the animals. The conc'ns of unchanged AZ in serum & tissue samples have been reported uncorrected and represent minimum values.

Results: Table 1 below shows the conc'ns of total RA in serum & tissues of male rats.

Table 1

Mean (± S.E.M.) Concentrations of Total Radioactivity in Serum and Tissues after Single Intramuscular Administration of ^{14}C-Aztreonam (50 mg/kg) to Groups of Three MALE Rats

Tissue	Concentrations of Aztreonam Equivalents in µg/g of Tissue or µg/ml of Serum at Time (hr)			
	0.25	2	8	24
Serum[a]	121 ± 10	17 ± 2.3	1.9 ± 0.1	0.5 ± 0.0
Adrenal gland	15 ± 4.2	3.1 ± 0.6	0.8 ± 0.1	0.3 ± 0.0
Bone	7.9 ± 2.4	0.4 ± 0.1	0.2 ± 0.1	0.1 ± 0.0
Bone Marrow	23.5 ± 3.3	2.4 ± 0.7	1.3 ± 0.7	0.2 ± 0.0
Brain	1.8 ± 0.2	0.4 ± 0.1	0.5 ± 0.3	0.1 ± 0.0
Eyes	9.1 ± 1.6	2.5 ± 0.4	0.4 ± 0.1	0.1 ± 0.0
Heart	18 ± 2.9	2.3 ± 0.4	0.6 ± 0.0	0.6 ± 0.4
Kidney	227 ± 35	47 ± 8.3	19 ± 3.0	14 ± 3.0
Large Intestine	16 ± 1.5	3.9 ± 0.9	112 ± 41	12 ± 1.3
Large Intest. Con.	0.5 ± 0.2	0.2 ± 0.1	117 ± 33	22 ± 2.3
Liver	158 ± 5.4	109 ± 14	51 ± 4.2	3.2 ± 0.2
Lung	31 ± 0.4	4.7 ± 0.8	0.9 ± 0.1	0.4 ± 0.1
Lymph Nodes	28 ± 6.9	5.5 ± 0.6	1.6 ± 0.3	0.4 ± 0.1
Meninges	64 ± 16	26.3 ± 6.8	2.4 ± 0.6	0.7 ± 0.0
Muscle	8.4 ± 0.8	1.9 ± 0.2	0.5 ± 0.1	0.2 ± 0.1
Muscle Inj. Site	546 ± 291	6.5 ± 2.2	1.6 ± 0.3	1.0 ± 0.3
Pancreas	8.9 ± 4.4	3.1 ± 0.5	0.8 ± 0.1	0.6 ± 0.2
Salivary Gland	21 ± 3.1	3.5 ± 0.5	0.9 ± 0.2	0.2 ± 0.0
Small Intestine	24 ± 1.2	215 ± 41	38 ± 18	0.3 ± 0.
Small Intest. Con.	10 ± 3.4	167 ± 13	142 ± 3.5	0.9 ± 0.2
Skin	28 ± 4.1	5.7 ± 0.9	0.9 ± 0.1	0.3 ± 0.0
Spleen	7.7 ± 0.3	1.8 ± 0.2	0.9 ± 0.3	0.5 ± 0.2
Stomach	19 ± 1.3	23 ± 6.1	1.6 ± 0.4	3.3 ± 1.8
Testes	7.2 ± 1.0	3.2 ± 0.5	1.1 ± 0.3	0.2 ± 0.0
Urinary Bladder	265 ± 135	865 ± 442	71 ± 17	1.4 ± 0.4

[a] Concentrations expressed as µg/ml.

Except the lymph nodes, similar results were obtained with the F rats. RA in the lymph nodes of F rats was higher than in serum after 2 hrs postdosing.

Average conc'ns of total RA in 6 tissues of interest to the applicant, relative to conc'n in serum, is shown in the Table 2 below.

Table 2

Tissue	Time (hr)							
	0.25		2		6		24	
	Male	Female	Male	Female	Male	Female	Male	Female
Serum	100	100	100	100	100	100	100	100
Kidney	189	190	274	203	987	796	2904	1238
Liver	132	145	633	392	2693	2261	670	604
Lung	26	32	37	38	45	93	81	107
Skin	23	20	32	39	46	45	58	51
Brain	2	2	2	2	25	6	24	8
Meninges	54	65	158	95	130	121	150	136

Conc'ns higher than in serum were seen in kidney & liver (excretory organs) and meninges.

Conc'ns of unchanged AZ as % of total RA in selected tissues are shown in Table 3 below.

Table 3

Tissue	Unchanged Aztreonam as Percent of Total Radioactivity Time (hr)			
	0.25	2	6	24
	MALE			
Serum	70 ± 6.0	50 ± 11	7.7 ± 2.4	2.3 ± 0.1
Kidney	51 ± 3.5	19 ± 7.4	3.9 ± 0.5	3.2 ± 0.5
Liver	34 ± 2.0	12 ± 2.8	3.4 ± 0.6	5.6 ± 1.3
Lung	75 ± 3.2	45 ± 2.3	9.0 ± 2.1	7.8 ± 3.9
Small Intes. Con.	53 ± 8.1	57 ± 4.1	46 ± 2.7	21 ± 1.6
Large Intes. Con.	21 ± 10	33 ± 10	57 ± 1.8	37 ± 1.4
	FEMALE			
Serum	68 ± 6.9	62 ± 6.7	5.8 ± 0.3	3.4 ± 1.4
Kidney	56 ± 1.9	19 ± 4.8	3.3 ± 0.2	3.7 ± 0.7
Liver	46 ± 11	14 ± 2.8	3.1 ± 0.7	2.5 ± 0.1
Lung	67 ± 2.0	34 ± 10	11 ± 3.9	7.7 ± 1.5
Small Intes. Con.	69 ± 2.0	62 ± 4.3	34 ± 5.9	27 ± 3.9
Large Intes. Con.	49 ± 5.5	42 ± 7.5	60 ± 1.5	31 ± 11

No major diff. between M & F were seen in tissue conc'n of unchanged AZ.

(Study #4)
Table 4

Mean (±S.E.M.) Concentrations (µg of Azthreonam Equivalents/g) of Total Radioactivity, Total Nonextractable Radioactivity, Azthreonam, SQ 26,992, and Other Extractable Metabolites (OEM) in Maternal Serum, Amniotic Fluid, Placentas, and Fetuses after a Single Subcutaneous Dose of ^{14}C-Azthreonam (150 mg/kg) to Groups of Four Pregnant Rats on Day 16 of Gestation.

Tissue	Time (hr)	Total Radioactivity	Total Non-extractable Radioactivity	Extractable Radioactivity		
				Azthreonam[a]	SQ 26,992	OEM
Maternal Serum	1	143.4 ± 13.7	12.8 ± 1.08	88.9 ± 12.3	24.7 ± 3.91	16.9 ± 6.09
Placentas	1	33.1 ± 2.07	2.65 ± 0.45	24.1 ± 2.03	2.74 ± 0.18	3.61 ± 1.79
Fetuses	1	2.14 ± 0.17	0.13 ± 0.04	1.10 ± 0.28	0.19 ± 0.06	0.73 ± 0.19
Amniotic Fluid	1	0.93 ± 0.14	0.04 ± 0.01	0.64 ± 0.09	0.10 ± 0.03	0.16 ± 0.02
Maternal Serum	2	44.6 ± 5.79	5.85 ± 0.50	26.5 ± 4.37	5.01 ± 1.22	7.18 ± 0.78
Placentas	2	15.5 ± 0.87	1.15 ± 0.30	8.44 ± 1.14	3.09 ± 0.41	2.79 ± 1.02
Fetuses	2	2.08 ± 0.11	0.13 ± 0.05	0.83 ± 0.19	0.19 ± 0.02	0.94 ± 0.22
Amniotic Fluid	2	0.88 ± 0.04	0.08 ± 0.01	0.51 ± 0.05	0.13 ± 0.03	0.1 ± 0.04
Maternal Serum	4	7.67 ± 0.30	2.87 ± 0.11	1.60 ± 0.34	2.65 ± 0.07	0.54 ± 0.16
Placentas	4	7.11 ± 0.57	1.05 ± 0.14	1.87 ± 0.09	3.49 ± 0.46	0.70 ± 0.08
Fetuses	4	1.92 ± 0.13	0.25 ± 0.05	0.88 ± 0.10	0.38 ± 0.03	0.41 ± 0.07
Amniotic Fluid	4	0.65 ± 0.06	0.06 ± 0.01	0.35 ± 0.04	0.11 ± 0.02	0.13 ± 0.02
Maternal Serum	8	3.47 ± 0.19	1.78 ± 0.07	0.10 ± 0.01	1.40 ± 0.21	0.20 ± 0.06
Placentas	8	4.98 ± 0.40	0.76 ± 0.27	1.15 ± 0.39	2.02 ± 0.35	1.04 ± 0.38
Fetuses	8	1.19 ± 0.07	0.11 ± 0.03	0.30 ± 0.03	0.48 ± 0.08	0.29 ± 0.02
Amniotic Fluid	8	0.69 ± 0.10	0.08 ± 0.01	0.28 ± 0.05	0.22 ± 0.03	0.12 ± 0.01

[a] Values reported are uncorrected for recovery of azthreonam in control spiked samples and represent minimum values (see text for details).

12-15-89

Results: There was a dose-dependent increase in the mean relative liver wts in both M & F. The relationship was more pronounced in the F, where the increase was sig. at all dose levels. When M & F were combined, the increases in rel. liver wt over that of controls were 20, 40 & 70% at 150, 600 & 2400mg/kg dose levels, respectively. The combined results also showed that there was no sig. increase in any of the enzyme activity measured or in microsomal protein conc'n at any dose level. On the contrary, there were sig. decreases in all these parameters at 2400mg/kg, when compared to controls and other dose gps.

Some differences among the sexes were reported. In M, there were slight increase in microsomal protein conc'ns in a dose-dependent manner. In F, the conc'n at the HD level was sig. lower than at the LD.

At all dose levels, aminopyrene N-demethylase activity tended to be much higher in the M than in the F. In the F, the enzyme activity was essentially similar at all dose levels. There was a decrease in enzyme activity in M at 2400 mg/kg dose level.

After a 2-mo. recovery period, all of the above changes showed a tendency to reverse. The increased liver wt was seen only at the HD level and the increase was not sig. Only the aniline hydroxylase activity was still sig. lower at the HD level, compared to controls.

7. Covalent Binding of ^{14}C-SQ & ^{14}C-Penicillin G to Human Serum Proteins

Method: Serum samples were prepared from blood obtained from 3 non-fasted M human volunteers. The labeled drugs in phosphate buffer (pH 7.4) were then added to the serum to final conc'ns equimolar to 10ug/ml of AZ. The incubation (aerobic) was carried out for 24 hrs at 37°C.

Results: The amounts of protein-bound RA for both compounds, whether expressed as percentages or conc'ns (wt equiv./ml serum or /mg protein) increased with incubation time. At 24 hrs, about 70% of AZ & 42% of penicillin G were bound to serum proteins. The binding was linear for penicillin G. For SQ, the rate of binding appeared to be slower during the first hr of incubation. When the conc'n of either drug was increased 10-fold, the binding also increased 10-fold. The exact nature of the binding remains unknown. It appeared that SQ was bound to proteins by covalent bonds to sulfhydryl gps in proteins. Penicillin G reacts with both sulfhydryl & amino gps under appropriate conditions (Wagner et al. J. Antibiotics 24: 647, 1971. Schneider et al. Nature, 208: 57, 1965).

8. A Pharmacokinetic Evaluation of AZ (#12)

Rat, mouse & monkey data have been reviewed before. (Pharm. Rev. of 5/12/81).

Gps of animals were treated IV (mice, rabbit), IM (squirrel monkeys), SC (rats, mice) or orally (mice, rats) with AZ, and serum levels & excretion were followed by microbiological assays.

The applicant stated that rabbits given AZ (25mg/kg), achieved peak serum conc'ns 10 min. after admin., followed by a linear fall in conc'n over a 2-hr period. "The mean concentrations of AZ in serum were 118 and 2.2 ug/ml at 10 and 120 minutes, respectively, after administration."

Toxicology

1. Acute IV Toxicologic & Pathologic Study in Mice

Material Tested: SQ-arginine blend

Species: CR CD-1 mice (M & F)

# of Animals: 30/gp for LD_{50}; 20/gp for maximal no-effect dose

Dose Levels: 800, 1000 or 1200mg/kg for "no effe dose"; 1450, 1750, 2100 or 2500mg/kg for LD_{50}

Route: IV

Period of Observation: 14 days

Results: Acute LD_{50}s (IV): M mice - 1785mg/kg; F mice - 1710mg/kg All deaths occurred within 5 min. except one F (2100mg/kg) at 9 days. Toxic signs immediately after dosing included ataxia, convulsions & collapse. Maximal "no effect" doses: 800mg/kg for F & 1000mg/kg for M.

Pathologic Evaluation: Only one animal (F given 2100mg/kg that died on 9th day) was examined histologically. This animal had epicarditis, focal hemorrhage in the lungs, severe edema of the skin, intestinal distention caused by edema, and severe membranous glomerulopathy.

All mice that died and all mice dosed with 1450 or 1750mg of SQ/kg were examined grossly. One M at 2100mg/kg had left testicular retraction; one F at 2100mg/kg & 2 F at 1450mg/kg had a slightly distended uterus filled with clear fluid. All other animals examined appeared normal grossly.

2. Acute IV Safety Test in Mice (Report #'s 29 & 30 in the NDA)

A number of single-dose acute tests have been compiled together under this reference. All these (except 1) are screening tests for various batches of the material, performed between 5/4/81 & 1/17/83. It was reported that at 1200mg/kg, there was no death. Ataxia lasting about a minute was seen immediately after dosing.

3. Acute IP Toxicity in Mice

The IP LD_{50} in mice was reported to be 4.65g/kg. The material tested has only been identified as "Aztreonam for injection, 20g/vial". It is not clear whether this was pure AZ or the arginine blend.

4. Acute Oral Safety Test in Mice

An oral dose of 5600mg/kg of "SQ 26,776 for injection" was nonlethal to mice (0/20 deaths). No overt signs were noted for 2 hrs post-dose or on the following days. The animals were observed for 8 days.

5. Subacute SC Screening Study in Mice

"Aztreonam for injection" in aqueous sol'n was injected SC at dose levels of 625-3500mg/kg to gps of F mice (5/gp). Observation period was until 3 days post-dose.

No deaths or overt signs of toxicity were reported. Two mice (3500 mg/kg) showed wt losses during the dosing period. Occasional blood at the injection sites in 5 min. to 2.5 hrs post-dose and fistulas at the injection sites in 1-5 hrs were reported.

6. Acute IV Toxicity of AZ in Mice with Hepatic or Renal Damage (#34)

Methods: Carbontetrachloride (CCl_4) and uranyl nitrate hexahydrate (UN) were used to damage the liver & kidneys, respectively in M mice. CCl_4 was administered orally and UN, IV. Organ damage by an LD_2 dose of CCl_4 or UN was established after 1 & 4 days, respectively. For LD_{50} determinations, the drug was therefore administered IV at these times.

Results: Table 5 below shows the results.

Table 5

Predose* (ml/kg) Organotoxic Agent	Saline (ml)	# Mice /Group	LD_{50} (mg/kg) (95% Conf. Limits)	Estimated LD_2 (mg/kg)
100% CCl_4, 5.0	-	15	2200 (2025-2400)	1625
-	5.0	15	1775 (1575-2000)	1275
0.2% Uranyl Nitrate, 5.0	-	20	2075 (1900-2250)	1375
-	5.0	15	1800 (1650-1975)	1400

*CCl_4 given orally 24 hrs prior to AZ injection.
UN given IV 4 days prior to AZ injection.

7. Acute IV Toxicologic & Pathologic Study in Rats

Material Tested: SQ-arginine blend in water

Species & # of Animals: CR CD rats; 5 or 10/gp

Dose Levels: 925, 1150 or 1425mg/kg for max. "no effect" dose; 1800, 2250 or 2800mg/kg for LD_{50} determination

Route: IV

Period of Observation: 14 days

Results: The LD_{50}s were reported to be 2200mg/kg in the M & 2390mg/kg in the F rats. The maximum "no-effect" doses reported were 925mg/kg for M & 1150mg/kg for F. The LD_{50} values in terms of arginine were 1540mg/kg for M & 1670mg/kg for F. Signs of toxicity included ataxia, slow or gasping respiration, convulsions, collapsing and death.

Pathologic Evaluation: Histologic exams were performed on 1 F rat given 2800mg/kg that died within 24 hrs after dosing and 1 M rat given 2250mg/kg that died within 48 hrs after dosing. The F was diagnosed to have perivascular edema of the lung, diffuse vacoular change & individual cell necrosis of the liver, focal necrosis of the mucosa, multifocal hemorrhage in the stomach & thymus, focal hemorrhage in the pancreas, severe tubular degeneration in the kidneys, and severe necrosis at the injection site (tail). The M had severe congestion in the lung & lymph nodes, vacuolar change in the liver & pancreas, severe tubular degeneration in the kidneys, multifocal hemorrhage in the thymus, and hemorrhage at the injection site (tail). All other animals examined were reported to be grossly normal.

8. Acute IP Toxicity in Neonatal & Young Adult Rats

"Aztreonam for injection" was given as single IP doses to neonatal (3-day-old) and young adult (42 to 45-day-old) rats. The animals were observed for 8-9 days. The acute LD_{50}s are shown in the Table 6 below.

Table 6

Species	Sex	Age	# Animals/Group	LD_{50} (C.L.) (mg/kg)
Rat	M,F	3	20	3625 (3225-4050)
Rat	M,F	42-46	10	2250 (2025-2525)

Signs of Toxicity: Wt loss, anoxia, decreased respiration rate, ataxia, tremors & death.

9. Acute IV Toxicity Study in Dogs (#38)

In this rather brief summary, the applicant stated that when SQ-arginine was given IV to beagle dogs (1/sex/gp), the max. "no effect" dose in unanesthetized animals was found to be 200mg/kg. At higher doses (400 & 800mg/kg) emesis was observed within minutes in most animals.

10. Five-week SC Toxicologic & Pathologic Study in Neonatal Rats (#43)

Material Tested: SQ-arginine blend

Species, Age & # of Animals: CR CD rat pups, 1 day old; 6/sex/gp

Dose Levels: 0, 75, 300 or 1200mg/kg (SQ)

Route, Frequency & Duration: SC; once/day, 7 days/wk for 5 weeks

Results:

- Clinical Observations: SC hemorrhage at the injection site was seen in 6/12 HD pups & 1/12 each in LD & control gps. Urine stain at urogenital area was seen in 1 HD, 1 MD & 1 LD pups. Alopecia at the injection sites has been reported in 2 HD & 1 MD pups. Two control pups had alopecia in the pelvic area.

- Ophthalmoscopic Exam: One HD pup (#55F) had pale retinal reflection under ophthalmoscopic light on day 31.

- Mortality: One MD pup (5M) was necropsied in moribund condition on day 14. One MD pup (58F) was reported missing from the litter. One LD pup (17M) was found dead on day 25 with blood around the nares. A HD rat (14M) was found dead on day 26.

- Body Wt: At the end of treatment, all treated M & F had slightly lower mean body wts than controls. The decreased gain was more pronounced in M than in F.

- Food Consumption: Not reported.

- Urinalysis: Mean urinary protein excretion was slightly increased among the MD animals. Urinary pH was slightly decreased in the HD gp.

- Hematology: HD rats had slight reduction in erythrocyte cts., elevated reticulocyte & total leukocyte cts., and polychromasia, anisocytosis & macrocytosis of erythocytes by the 5th wk. No other remarkable changes reported.

- Clinical Chemistry: One MD rat (57F) had sig. elevated CPK value. All other parameters were within normal range.

- Organ Wt: Mean rel. liver & spleen wts were higher in the HD animals when compared to controls. All other organ wts were similar in all gps.

- Gross Pathology: Slightly enlarged liver & kidney were reported in 1 HD F & 1 M. Injection sites of all HD rats showed moderate or severe redness, occasionally with blood clots. One HD M had distended abdomen filled with clear fluid.

- Histopathology: Marked hemorrhage and marked subacute inflammation at the injection sites were seen in all HD rats. Minimal irritation at the injection sites of the MD & LD animals has been reported. No irritation was present in saline controls. Tiny foci of extramedullary hematopoiesis was seen in many livers from all groups but the incidence was more in the HD & MD groups (7/12) than in the control group. The lungs of several test and control rats showed mild peribronchial accumulation of lymphocytes and congestion and/or hemorrhage. Very mild to mild subacute interstitial inflammation has been reported in a

few animals of all treated groups but none in the control. (Other occasional lesions seem to be incidental.) Slight to marked dilation of renal pelvis were seen in 5/12 HD, 2/12 MD, 3/12 LD & 1/12 control group. The incidence in the HD group was significantly greater than in the controls.

11. Two-week Oral Toxicity Study in Rats

Material Tested: SQ-arginine blend, 20% aqueous sol'n

Species & # of Animals: CR CD rats; 6/sex/gp

Dose Levels: 0, 300, 1200 or 4800mg/kg/day

Route, Frequency & Duration: Oral (gavage); 2x/day, 5 hrs apart, for 13-20 days

Results:

- General Observations: All drug-treated rats had soft stool from day 2 and by the 2nd wk, distended or pendulous abdomens, till the end of the experiment. The magnitude of these changes was dose-related. Slight excessive salivation of short duration immediately after dosing, was noticed in 2 HD rats on day 5. One HD rat (112F) had a small area of gold-brown pinpoint foci on the retina of the left eye on day 14, and 1 rat at the LD had diffuse retinitis in the right eye on day 14.

- Body Wt: Similar gains in all gps.

- Food Intake: Normal in all gps.

- Mortality: None

- Ophthalmic Exam: Reported with the general observations.

- Urinalysis: Dose-related moderate to marked decreases in Na excretion & urinary pH were found in MD & HD animals.

- Hematology: Unremarkable; a differential blood count has not been done.

- Clinical Chemistry: MD males had significantly higher BUN than the controls, but the group means (M & F combined) were similar. Decreased serum calcium was found in the MD & LD groups. In the HD group, slightly increased CPK values, slight decrease in serum glucose (F), a very slight increase in serum GPT (F), and a slight increase in serum magnesium have been reported.

- Organ Wt: The mean rel. liver wts of the MD & HD animals were slightly but sig. lower than control values. No other remarkable changes were noted.

- Gross Pathology: The only sig. lesion reported was dilation of the cecum in the HD (12/12) & MD (12/12) animals. Also, smaller prostate gland & seminal vesicles were noted in 3 HD & 2 MD M.

- Histopathology: No changes were seen microscopically in the dilated ceca or in the grossly smaller prostate gland & seminal vesicles. Mononuclear cell infiltration in the liver, congestion in the heart & lungs, renal tubular calcification & ultimobranchial rests in the thyroid gland were present in treated as well as control gps. Chronic nephritis was reported in 1 HD F. Renal tubular dilation was noted in 2 MD M.

12. Two-Week Oral Toxicity Study in Dogs

Material Tested: SQ-arginine blend, 20% aq. sol'n

Species & # of Animals: Beagle dogs; 2/sex/gp

Dose Levels: 0, 100, 400 & 1600mg/kg/day

Route, Frequency & Duration: Oral (gavage); 2x/day (5 hrs apart) for 15-20 consec. days

Results:

- General Observations: soft or loose feces continuously throughout the dosing period in HD dogs, occasionally in MD, and once or twice in 2 LD dogs. Emesis was observed in one HD dog on day 1. Ond HD dog had mucus in feces (day 11), 2 MD dogs (305M & 308F) had mucus in feces on serveral occasions and 1 control dog (316F) had mucus and blood in feces on several occasions.

- Mortality: None

- Ophthalmoscopic Exam: No unusual findings reported.

- Body Wt: Similar in all gps

- Food Intake: Not reported.

- Urinalysis: Moderate decrease in urinary excretion of Na in the MD & HD gps, lower urinary pH, & higher water intake were reported.

- Hematology & Clinical Chemistry: Slight increase in GPT, BUN & PPT (gp mean values) reported for the HD gp, compared to pre-test values. No other sig. changes were noted.

- Organ Wt: No sig. organ-weight differences were noted between control & treated gps. Slightly lower mean rel. testes wt than the control value was reported for the HD group.

- Gross Pathology: Unremarkable

- Histopathology: No treatment-related lesion was apparent. Congestion & mineralization in collecting tubules of the kidney, mucosal hemorrhage in the large intestine, chronic inflammation & hepatocyte vacuolation in the liver, chronic multifocal inflammation in the lung, and hemosiderosis in the liver & spleen of the HD animals (usually involving 1 animal) were noted. These and other lesions noted are considered by the applicant to be incidental.

13. Six-Month SC Toxicity Study in Rats

Material Tested: SQ-arginine blend

Species & # of Animals: CR CD rats, 30 (15/sex/gp) & 10 (5/sex/gp' recovery animals.

Dose Levels: 0, 150, 600 or 2400mg/kg of SQ/day

Route, Frequency & Duration: SC; 2x/day for 6 mos. A satallite gp was dosed at the same schedule, but allowed to recover for 10 wks before sacrificing.

Results:

- General Observations: Slight to marked irritation at injection site with "gelateneous subcutaneous mass or swelling", blue-black discoloration, thickened or necrotic skin, serous discharge, cutaneous fistulas, scab & scar formations, alopecia and/or hemorrhage was reported in the rats of the HD group. "Some instances of focal ulceration and/or hyperesthesia" were also reported by the applicant. Ischemic retinal reflection in 1 M & 1 F was reported. With the exception of the last observation, all other lesions were also seen in MD animals, but with lower incidence & less severity. Among the LD animals, very slight irritation at injection sites with gelatinous subcutaneous mass or swelling, alopecia and/or hemmorhage were noted. Slight irritation also noted among the control animals.

- Mortality: One HD, 1 LD & 1 control rats died during the dosing period. One recovery animal from the HD gp also died on day 58 post-dose.

- Body Wt: Moderate decrease in body wt gains of M and moderate increase in body weight gains of F were reported for the HD animals.

- Food Intake: Not reported.

- Ophthalmoscopic Exam: At wk 25 exam time, slight retinal ischemia, focal retinitis or exophthalmos & granular lens opacity was reported in 3 HD rats. One MD rat had retinitis and another had diffuse retinal degeneration at wk 25. Marked retinal eschemia was reported in 1 LD rat, focal retinitis in another LD rat and patchy retinal degeneration in 1 LD rat at wk 25. In the control animals, diffuse retinitis in 1 animal & focal retinitis in another animal were reported. A few rats

with conjunctivitis was reported in all treated gps. All these lesions were seen in all gps at one time or another and the no. of animals with lesions was the same in the HD & control gps (10).

- Hematology: Dose-related decrease in Hgb, Hct & erythrocytes were reported (moderate at the HD & slight at the MD). Marked increase in reticulocytes (15x the control values) and moderate increase in platelet counts in the HD group, and slight increases in reticulocyte & platelet cts. have been reported in the MD group. Moderate to marked leukocytosis was noted in the HD group. Polychromasia and/or macrocytic anisocytosis of erythrocytes were noted in all HD animals and in some MD animals. Slight decrease in bone marrow lymphocytes were noted in the MD & HD group animals. None of these changes were noted in the LD or control animals. Bone marrow morphology & m/e ratio were not affected.

- Clinical Chemistry: Slight but sig. decreases in serum urea nitrogen, creatinine, glucose, cholesterol, total proteins, GPT, alkaline phosphatase, LDH & calcium in the HD gp, and slight but sig. decreases in cholesterol & GPT in the LD animals have been reported. Occàsional slight decreases in GPT values were noted in LD animals.

- Urinalysis: Water consumption & urine output moderately increased in the HD animals and slightly increased in the LD animals. Urinary protein conc'n was significantly higher among the HD animals at all measurement times and on wk 25 in the MD animals and LD animals, when compared to controls. slight but sig. increase in urine urobilinogen conc'n in M of the HDgroup was noted. Slight increase in urine specific gravity in the HD group has been reported.

- Organ Wt: Increases in gp mean abs. & rel. organ wts have been reported for liver, kidneys, heart & spleen in the MD & HD animals. Increases in the heart & liver wts have been reported for the LD animals also. Additionally increases in abs. gp mean wts have been reported for adrenals (all treated M & HD F), pituitary (HD & LD M, MD F) and ovary & thyroid (HD F). Significant increases in rel. wts were noted in adrenals (all treated M), brain (HD M), pituitary (all treated M), seminal vesicles (HD M), ovaries (HD F) & testes (HD M). The relative wts of brain in HD F, pituitary in LD F and thymus in MD & HD animals were significantly reduced from controls.

- Gross Pathology: Bluish coloration at injection sites (1 HD M); enlarged & pale kidneys (1 HD F); slight to moderate dilation of cecum (29 HD, 15 MD & 4 LD animals); slight splenomegaly (1 HD M & 1F) and sparse hemorrhage in thymus (1 HD M) have been reported.

- Histopathology: Animals sacrificed at the end of treatment period showed sig. increases in the severity & incidence of SC hemorrhage, hemosiderosis & proliferation of granulation tissues (injection sites) in HD & MD animals & LD M; vacuolation in the renal tubular epithelial cells in HD & MD animals & in 1 LD M; renal tubular dilation and extramedullary hematopoiesis in livers of HD M; focal infiltration of

mononuclear cells in kidneys of HD animals; [illegible]erosis in liver cells of HD F and in small intestines of HD [illegible]creased # of corpora lutea & infiltration of mononuclear cells in sclera in HD F. Significant increases in severity & incidence were also seen in bone-marrow leukoblastic & erythroblastic hyperplasia, and in apical vacuolation in mucosal epithelial cells of the urinary bladder of HD & MD animals. Brown pigmentation in lymph nodes in HD M, splenomegaly and infiltration of mononuclear cells in lacrimal glands of HD animals were also noted.

- Recovery Animals: Most of the hematological & clinical parameters either returned to normal or were showing improvements. Organ wts of animals showed increases in the wts of adrenal glands, heart, kidneys, liver, seminal vesicle & thyroid gland of HD M. Some increases in heart wts of LD M and increases in liver wts of MD & LD M have also been reported.

  Histologically sig. lesions in this gps of animals included hemosiderosis in the subcutis (injection sites) of HD & MD animals, in the liver cells of HD F and in the small intestines of HD M; many regressing corpora lutea in HD F and sig. increases in inflammatory reactions (nephritis & infiltration of monomuclear cells) in the HD animals. The severity & incidence of the vacuolation in renal tubular epithelial cells of HD & MD animals were significantly decreased.

- Serum Conc'n & Urinary Excretion of AZ: Serum & urinary conc'ns of AZ were determined in the samples obtained during the 6-month toxicity studies. The highest mean serum conc'n in all 3 dose gps was found at 0.5 hrs after the 2nd dose. Serum conc'ns at the 16th & 23rd wk were similar at each dose level, indicating no cumulation or any change in the rate of elimination of AZ in the rats during treatment. Values at the 5th wk were generally lower due to greater breakdown of AZ. At the LD (150mg/kg) the serum conc'n on wk 23 was 120ug/kg & at the HD (2400mg/kg) the value was 608ug/kg for the F. The M had similar values. Average daily urinary excretion of AZ in rats ranged from 47% (LD F) to essentially 100% (HD) of the admin. dose. Greater amount of urinary recovery of AZ was found in M than in F. No statis. sig. differences in urinary excretion were found during the 4th, 15th & 24th wks of the study.

- Ultrastructural Evaluation:

  - HD Group:

    - Liver: The applicant stated that the liver of all rats at the HD (3/sex) exhibited normal cytostructure. 3 rats showed slight increase in fesiculated SER. The Golgi vesicles appeared mostly swollen and contained electron dense material. Single membrane bound peculiar peribiliary residual bodies were seen in all livers of this gp. These bodies were more numerous & bigger in size in the F than in the M.

- Kidneys: The renal tubular lesions (vacuolation & abundance of heterolysosomes) appeared to be less severe in this gp than that seen in animals without recovery period. The membranes of the glomerular capillaries and the podocytes were normal.

- MD Group:

  - Liver: The ultrastructure "appeared normal" and showed a slight increase in the SER vesicles in 2 F & 1 M.

  - Kidneys: The applicant stated that the size & no. of lysosomes were mildly increase in the renal tubules of 2 F & 1 M. Additionally, vacuolation of the tubules was reported in 1 F (8121-204-F2). Podocytes appeared swollen & enlarged in 1 M (8121-091-M2). Glomerular capillaries appeared normal.

- LD Group: The applicant stated the "the liver and kidney ultrastrcuture did not show any significant change and appeared generally normal in both the female and male rats examined in this group." A few very small lysosomes were seen, particularly in the renal proximal tubules, but this was considered normal.

14. Six-Month SC Toxicity Study in Dogs

Material Tested: SQ-arginine blend

Species & # of Animals: Beagle dogs; 6 (3/sex) and 2 (1/sex) recovery animals

Dose Levels: 0, 50, 200 & 800mg of SQ

Route, Frequency & Duration: SC; 2x/day in divided doses for 6 mos.

Results:

- General Observations: Slight irritation at injection sites, with large, gelatinous SC masses & slight swelling, thickened skin, occasional bleeding & slight drug leakage during wks 1-23 have been reported in the high-dose group. After wk 23, marked irritation at the injections sites with swelling, pain, bleeding, serous discharge & more profuse drug leakage were noted. Tissue overlying injection sites was very hard on palpation. Two dogs in poor condition were sacrificed. One (703M) had a large hematoma on the rt. lateral thorax during the 2nd wk post-dose. Repeated emesis was noted in these dogs.

  In the MD gp, small, gelatinous SC masses at the injection sites that tended to disperse quickly were reported. Slight irritation with slight swelling & bleeding were noted in this gp of animals.

  In the LD gp, very slight irritation at the injection sites were reported.

Other observations appear to be incidental and no dose-related effects were noted.

- Body Wt: Similar gains in all gps.

- Food Intake: HD F had lower intake compared to controls at wks 26, 27 & 28.

- Mortality: Two dogs (702M & 703M) were sacrificed in poor condition on days 109 & pose-dose day 9, respectively.

- Ophthalmoscopic Exam: Not performed.

- Hematology: The 2 HD dogs that were sacrificed in moribund condition had some drastic changes in hematology parameters. Dog 702M showed increased erythrocyte osmotic fragility during wk 13 & was severely anemic by wk 14. There were no circulating reticulocytes in this dog, and there was a marked absence of erythroid cells in bone marrow. Dog 7093M that showed moderate bleeding from the injection site throughout the dosing period, showed marked & sig. decrease in Hct, Hgb & erythrocytes and sig. increase in total leukocytes & reticulocyte cts; platelet cts. were normal. Dog 707F of the HD gp showed a slight increase in osmotic fragility of erythrocytes. HD & MD M dogs showed very slight decrease in Hgb, Hct & erythrocyte cts. HD dog 707F showed a slight increase in erythrocyte osmotic fragility at wk 26.

- Clinical Chemistry: Dog 702M (sac'd early), prior to necropsy showed slight to moderate increases in serum urea nitrogen, total protein and GOT, and marked increases in serum GOT & LDH. Dog 703M, prior to necropsy, showed sig. decreases in serum K & calcium conc'ns and significant increases in CPK & GOT values.

  The HD dogs showed slight decreases (mean values) in total proteins, creatinine & serum glucose, and very slight increase in serum Na, cholesterol & LDH.

  MD dogs had a slight decrease in serum total proteins. Other individual & gp changes were minor and no dose- or time-relationship was observed.

- Urinalysis: Slight increase in specific gravity & a slight decrease in pH were noted at the HD.

- Organ Wt: Sig. increases in the abs. & rel. liver wts of HD & LD F and in rel. kidney wt of HD F were noted. Absolute liver wt of HD M was also significantly increased. A sig. decrease in the rel. heart wt of HD & MD M, an increase in the abs. testicular wt of LD M were also reported. Liver wts were also increased in the LD & MD as compared to controls, but the increases were not significant.

- Gross Pathology: Cavitation & hemorrhage at the injection sites.

- Histopathology: The following dose-related changes were noted: increase in severity & incidence of lesions at injections sites (necrosis, fibrosis, hemorrhage & inflammatory cell infiltration); increase in severity of cytoplasmic change in the liver & increase in incidence of cytoplasmic vacuolation in renal tubules.

  Lesions at the injection sites were centered primarily in subcutis and were characterized by necrosis often accompanied by hemorrhage, mixed inalammatory cell infiltration & fibrosis. Tissue necrosis was not present in LD & control gps.

  The cytoplasmic change in the liver was characterized by reduced cytoplasmic density of hepatocytes (glycogen accumulation?) in all gps, but the severity of the change was dose-related and was most prominent in F. In severely affected livers, this change was associated with cellular hypertrophy. Necrosis was reported in 1 HD liver.

  The renal lesions consisted of cytoplasmic vacuolation of tubular cells primarily of the collecting ducts; the severity was minimal and, although these lesions were present in control dogs also, the incidence was greater in the the treated gps, particularly among the F. Nephrocalcinosis was reported in 4/6 HD, 3/6 MD & LD and 2/6 control animals.

  Other lesions reported are hemosiderosis & extramedullary hematopoiesis in the spleen, bone marrow hypoplasia, epicarditis & lymphocytic infiltration in the heart and testicular degeneration. Single instances of these lesions were reported only in the HD gp.

  Erythrophagocytosis in peripheral lymph nodes was noted in animals from all gps. Also estrus-related changes were seen the the F.

- Recovery Animals: Since there were only 2 (1/sex) in each gp, the applicant stated that no definitive statement could be made as to reversibility of the liver & kidney changes seen in the treated animals.

  However, the lesions at the injection sites appear to have reversed in post-dose animals.

- Ultrastructure Evaluation: (Electron Microscopy)

  - Control Animals: In light microscopy, the hepatocytes of controls (3 examined) appeared normal with clear cytoplasm. Electron microscopic exam revealed heavy deposition of glycogen with scanty SER pushed to the periphery of the cells. RER was hardly noticeable.

    The kidneys of these dogs were normal. The renal tubules showed intact cytostructure with a regular pattern of apical vacuoles, mostly located near the brush border, and lysosomes of moderate size.

  - HD Animals: Five were examined. In the livers of these animals, enlarged hepatocytes with marked deposition of glycogen were seen,

and the SER was markedly increased in M & moderately increased in F. The mitochondria appeared normal but somewhat smaller. Both the mitochondria & the proliferating SER were pushed to the periphery of the hepatocytes by the glycogen deposits. Lipofucsin-like pigment containing fat droplets was also seen scattered in the hepatocytes.

The kidneys showed marked lysosomal activity, and numerous enlarged apical vacuoles were present.

- Recovery Dog: Only one was examined. Sig. increase in the RER in hepatocytes was noted. Glycogen deposition was still dense in different zones of the hepatic nodule. Lipofucsin pigment was also still noticeable, though less than seen in the dogs sacrificed at the end of treatment.

  The renal cytostructure was "mostly normal". The lysosomes and apical vacuoles were few and in the "normal range".

15. Mutagenicity Testing: Mouse Bone Marrow Cytogenetic Assay

    Lab Perf. Study: Litton Bionetics, Inc., Kensington, MD

    Material Tested: AZ

    Species & # of Animals: 8 CR CD-1 mice

    Dose Levels: 0, 400, 1200 or 3600mg/kg

    Route, Frequency & Duration: SC; acute - a single dose; subshcronic - one dose/day for 5 days

    Method: Gps of animals were given the drug SC at appropriate dose levels & 3 hrs prior to killing were injected IP with 4mg/kg of colchicine. Negative controls and treatment gps were killed at 6, 24 or 48 hrs after dosing and bone marrow slides were made. Bone marrow metaphase chromosomes were then examined for structural changes & rearrangements. Triethylenemelamine (TEM) wasd used as postive control, and these animals were killed at 24 hrs after dosing.

    Results: The range of structural aberration frequency in the negative control animals was low (0-0.005). Similar results were obtained with the 3 dose levels of AZ. TEM, on the other hand, increased the frequency of structural aberrations. AZ was therefore non-mutagenic in this assay.

16. Mutagenicity Testing of AZ (Performed by Squibb, Italy)

    English translation of these 2 Italian studies has been provided.

    a. Ames Test: (identified as CRF Study 364/M)

       Methods: The method of Ames et al. was used. Salmonella typhimurium

strains TA 98, 100, 1535, 1537 & 1538 were used. The assay was carried out in both the presence & absence of rat liver S-9 activation system.

Results: The material did not induce any sig. increase in retromutants in the strains in either the presence or absence of metabolic activation.

b. Mitotic Gene Conversion in S. cerevisiae (D4)

The method developed by F.K. Zimmerman (Mutation Res. 31: 73, 1975) was used.

AZ, in both the presence & absence of metabolic activation was found not to induce any sig. increase in the frequency of gene conversion in S. cerevisiae (D4) strain when compared to controls.

17. Mutagenicity Testing Measuring Chromosome Aberration in Human Lymphocytes (In Vitro)

Lab Perf. Study: Litton Bionetics, Inc., Kensington, MD

Material Tested: AZ

Methods: Human lymphocytes in culture were treated with PHA (phytohemoglutinin) and incubated for 24 hrs. At this time, sol'ns of test compound, EMS (ethylmethanesulfonate) or solvent was added, and the lymphocytes further incubated for 22 hrs. with or without S-9 fraction (rat liver). The cells were then resuspended in medium containing colcemid (0.1mg/ml), incubated for a further 2 hrs and checked for chromosomal aberrations after fixing and staining.

Results: There was no sig. increase in chromosome aberrations over the background in the presence or absence of metabolic activation.

At 50 & 70ug/ml dose level, AZ appeared to have a suppressive effect on mitosis in this system.

18. SC Ototoxicity Study in Neonatal Rats (58)

Methods: Three gps of 16 neonatal rats each were given SQ (AZ) SC at total daily doses of 150, 600 or 2400mg/kg, from day 10 thru 16, post-natally. Streptomycin (400mg/kg) was given to the positive control gp and the negative control gp received 0.9% saline on the same schedule.

Results: Streptomycin was highly ototoxic, producing morphologic changes in the sensory nerve endings of the auditory & vestibular portions of the ear. In the cochlea, all of the hair cells and most of the supporting elements in the organ of Corti were missing. Atrophy of spiral ganglia was evident. These changes correlated well with severe impairment of auditory function in these neonates. SQ-treated neonates did not show any signs of ototoxic effects.

19. Acute Toxicity Studies in Rat & Mice

Lab Performing Study: NRI Life Sciences, Kanagawa, Japan

These studies were performed in Japan as requirement for marketing of the drug in that country. The results are summarized in Table 7 below:

Table 7

Species	Route	Sex	LD50 (mg/kg)	95% confidence limit (mg/kg)
Rat	P.O.	Male	>10000	
		Female	>10000	
	S.C.	Male	3578	(3445 - 3716)
		Female	3154	(2973 - 3346)
	I.V.	Male	2882	(2752 - 3019)
		Female	3149	(2998 - 3307)
	I.P.	Male	2549	(2388 - 2721)
		Female	2964	(2727 - 3221)
Mouse	P.O.	Male	>10000	
		Female	>10000	
	S.C.	Male	3906	(3592 - 4247)
		Female	5368	(4809 - 5993)
	I.V.	Male	1963	(1823 - 2115)
		Female	2068	(1929 - 2217)
	I.P.	Male	2897	(2705 - 3102)
		Female	3722	(3424 - 4046)

20. Five-Week IV Subacute Toxicity Study in Rats

Lab Perf. Study: NRI Life Sciences, Kanagawa Japan

Material: SQ-arginine blend

Species & # of Animals: SD rats; 40 (20/sex); 32 (16/sex) in 100mg/kg gp

Dose Levels: 0, 100, 270, 750 or 2000mg/kg/day

Route & Duration: IV; 35 consecutive days

Recovery Period: 35 days after cessation of treatment

Results:

- Clinical Signs: None reported in the control & 100mg/kg gps. Reported at 270mg/kg were yellowish & soft feces in 2 M on days 4-8 and soft feces in 2 F on day 30. At 750mg/kg, soft & yellowish feces sometimes with mucus were observed in 7/sex rats, but after 16 days these findings were not present. Also 1 M of this gp had convulsions after dosing & died immediately on day 34. The 2000mg/kg gp showed the above fecal abnormalities: swelling of the extremeties & face, reddening of the extremeties & pinna in almost all cases, sedation or ventral decubitus in may cases, labored breather, a fall in body temp., convulsions, piloerection, loss of lustre of the hair and emaciation.

- Mortality: There were 18 deaths (1 MD M, 8 HD M, 9 HD F) during treatment.

- Body Wt: Slight increase in body wt gains in 100-750mg/kg M, and F at the HD (2000mg/kg) showed sig. increase in body wt (15%) as compared to controls.

- Food Intake: No sig. findings

- Water Intake: Increase in the HD M & F was reported.

- Ophthalmoscopy: No abnormal findings

- Urinalysis: Slight decrease in urine vol. in the HD F and an i eased K excretion in the MD F were reported.

- Hematology: Slightly dec eased erythrocyte & Hgb cts., a decrease in lymphocyte %'s, elevation of MCV & MCH and a rise in the segmented leukocytes were noted in the HD M. In the HD F, slight decrease in Hct & Hgb and a tendency for increase in lymphocyte %'s were noted in all treated gps, but variations were slight and dose relationship was marginal.

- Clinical Chemistry: In the M & F, a decrease in GOT & GPT values was seen in a dose-related manner. No sig. changes were observed in the 100 & 270mg/kg gps. In the HD F, variations in blood sugar & creatinine values were observed sporadically.

- Gross Pathology:

  - Dead Animals: The dead 750mg/kg rat showed enlargement of the liver & kidneys and discoloration of the kidneys. In the 2000mg/kg M, enlargement of the liver (8), enlargement of the kidneys (6) & enlargement of the cecum (3) were noted. Single instances of pale kidneys, atrophied spleen, reddish discoloration of the thyroid, dilatation of the rt. ventricle of the heart, hyperemia in the glandular stomach and hypertrophy of the adrenals were reported.

In the HD females, dark reddish discoloration of the lungs (7), enlargement of the liver (8), enlargement of the cecum (4), enlargement of the kidneys (6), pale kidneys (2) & adrenal hypertrophy (1) were reported.

- Surviving Animals: Cecal enlargement was found in one 270mg/kg M, all 750 & 2000mg/kg M, 3/10 750mg/kg F and 4/10 2000mg/kg F. Pelvic dilation in kidneys in one 270mg/kg M was reported. Other lesions noted were enlargement of kidneys, reddish discoloration of kidneys, atrophy of the testes, epididymides & prostates in 1 or 2 animals in the 750 and/or 2000mg/kg gps.

- Organ Wt: Dose-related increase in abs. & rel. wts of the cecum was seen in the groups that received 270mg/kg or more of drug (M) and in all F gps. Increases in abs. liver wts in all rats & increased rel. liver wts in F of 270mg/kg or more have been reported. At the HD level, increased kidney, spleen & thymus wts were noted.

- Histopathology:

  - Surviving Animals: Moderate centrilobular hypertrophy of hepatocyte was seen in all animals of the 750 & 2000mg/kg gps. Two of the 10 F of the 270mg/kg gp showed slight hepatocyte hypertrophy. No fat deposits or increased glycogen was noted. In the recovery group no hypertrophy was present in any animal.

Slight to moderate vacuolation of tubular epithelium of kidneys was seen in all 750 & 2000mg/kg gp animals. Vacuoles were not due to fat deposits or glycogen because sudan III staining & PAs tests were negative. Additionally, regeneration of proximal tubular epithelium and moderate hypertrophy of Bowman's capsule and thickening of basement membrane were seen in 2/6 M at 2000mg/kg. Dilatation of the renal pelvis & interstitial cellular infiltration (considered accidental) were reported in 1 M at 270mg/kg. In the recovery animals, no tubular epithelium vacuolation was noted. Thickening of Bowman's capsule & basement membrane of tubular epithelium, and regeneration of proximal tubular epithelium were still present in 4/6 HD M & 2/6 HD F.

In the spleen, slightly increased extramedullary hematopoiesis (red pulp), hyperplasia of the white pulp & hyperactivity of the germinal center were reported in all M & F of the 2000mg/kg gp. In the F some animals of the 750mg/kg were also affected. In the recovery animals, only a few F showed these lesions, while all M had fully recovered.

Other lesions reported were isolated cases and did not appear to be treatment-related.

- Dead Animals: Findings common to the dead cases were centrilobular vacuolation of hepatocyte and vacuolation of the proximal tubular epithelia of the kidneys. Congestion of the lungs was observed in both M & F. M of the HD gp also showed interstitial hemorrhage of the kidney, involution of red pulp of the spleen, congestion of the thyroid & hyperemia in the mucosa of the glandular stomach. In the F of the 2000mg/kg gp, congestion of the thyroid was reported in 1 case.

- Electron Microscopic Evaluation: Liver & kidneys from 2 each of control & HD F from both test & recovery gps were examined.

  - Liver: An increase in the lipid droplets and lysosome-like granules and atypical mitochondrias were seen in the treated livers when compared to the controls Also, "Kupffer cells had many large-sized vacuoles of relatively low density and occasionally showed myelin-like structures."

    In the recovery animals, these changes were seen only occasionally and the severity was slight. Kupffer cells not affected.

  - Kidneys: Myelin-like structure was seen in the glomerular epithelium. The nucleus of mesangium cell was darker than in the control group.

    Compared to controls, an increase in lysosome-like granules & vacuoles in cytoplasm in proximal tubular epithelium. No lesion seen in distal tubular epithelium.

    In the recovery animals no vacuoles seen. Other lesion reported above were seen only occasionally and a recovery was apparent.

21. IV Two-Generation Reproduction Study in Rats

Lab Perf. Study: NRI Life Science

Material Tested: SQ-arginine blend

Species: SD rats
# of animals: See table below.

	Dose (mg/kg)			
	Control	100	250	750
# of mated F rats	37	37	37	41
# of pregnant dams	35	36	33	34
# of dams used C-section	23	24	22	22
# of dams in lactation study	12	12	11	12
# of dams delivered F_1	12	12	11	12

Dose Levels: 0, 100, 270 or 750mg/kg

Route & Duration: IV; days 7-17 of gestation

Results:

Effects on F0 Dams: No deaths were observed in any gps. No abnormality was reported in any dams during gestation period, delivery or lactation period. In the HD group, an increase in water consumption was reported during the gestation period.

Gross Pathology did not reveal any sig. abnormalities. Cecum wt increases (abs. and/or rel.) were seen in some dams (all gps) necropsied on day 20 of gestation, as well as those necropsied on day 21 of lactation.

Reproductive Parameters:

Table 8

	SQ 26,776 (mg/kg)			
20-Day Necropsy	Control	100	250	750
Pregnancy rate (%)	100	100	100	100
# Corpora Lutea (mean)	15.3	16.4	16.0	16.7
Preimplantation loss (%)	5.7	16.8	8.2	10.6
# Implantations (mean)	14.5	13.7	14.7	15.0
Resorptions (%)	5.7	3.0	8.7	3.3
Dead fetuses (%)	0	0	0	0
# Live fetuses (%)	13.7	13.2	13.4	14.3
F1 Fetal Observations (20-day cesarian)				
M/F ratio	0.91	0.85	1.13	1.21
Body length (mm) - Male (mean)	39.3	39.4	39.7	39.1
Female "	38.5	38.5	38.7	38.2
Tail length (mm) - Male "	14.0	14.2	14.2	13.9
Female "	14.0	14.1	14.0	13.9
Body weight (g) - Male "	3.86	3.91	3.94	3.80
Female "	3.66	3.64	3.67	3.57
Adhesion of placenta	1	0	3	0
Subcutaneous hemorrhage	0	0	1	0

Abnormalities: Visceral, skeletal or external exam did not reveal any gp differences in abnormalities.

Reproductive Parameters on Dams Allowed to Give Birth: Shown in Table 9.

Table 9

	SQ 26,776 (mg/kg)			
	Control	100	250	750
# of Litters	12	12	11	12
# of Implantations (mean)	12.9	15.3	15.0	14.6
Mean length of gestation (days)	21.7	21.8	21.8	22.1

Observations on F_1 Offspring:

Table 10

Parameters	SQ 26,776 (mg/kg) Control	100	250	750
# of Live Fetuses:				
Postpartum day 0 (mean)	12.3	13.4	13.6	13.3
1 "	11.8	13.4	13.6	13.1
4 "	11.8	13.4	13.6	11.5
21 "	9.1	9.2	10.0	8.3
Sex ratio (M/F)	1.18	1.09	0.81	1.00
Live birth index (%)	95.5	87.5	90.9	91.4
Viability index (%)	95.3	100.0	100.0	86.2
Weaning index (%)	100.0	100.0	100.0	100.0
# w/malformation: agnesis of the sacrococcygeal vertebrae	0	0	1	0

Reproductive Parameters - F_1 Dams & F_2 Fetal Observations

Table 11

	SQ 26,776 (mg/kg) Control	100	250	750
# of litters (F_1)	14	11	9	12
# corpora lutea (mean)	15.5	15.1	15.9	15.7
Preimplntation loss (%)	8.8	7.2	15.4	5.3
# implantations (mean)	14.1	14.0	13.4	14.8
Resorptions (%)	3.0	1.3	4.1	1.7
Dead fetuses (%)	0	0	0	0
Total deal implantation (%)	3.0	1.3	5.0	1.7
# Live fetuses (mean)	13.7	13.8	12.8	14.6
Sex ratio (M/F)	1.16	1.30	1.25	0.80
Mean body length (mm) - Males	39.2	39.5	39.4	39.5
Females	38.3	38.8	38.4	38.7
Mean tail length (mm) - Males	14.0	14.2	14.0	14.3
Females	14.1	14.2	14.0	14.2
Body weight (g) - Males	3.68	3.75	3.74	3.85
Females	3.52	3.58	3.52	3.63
# fetuses w/malformations	0	0	0	0

Reproductive Parameters - F_1 Dams Allowed to Give Birth and & F_2 Fetal Offspring Observations

Table 12

	SQ 26,776 (mg/kg)			
	Control	100	250	750
# of Litters (F_1)	8	7	7	7
Mean length of gestation (days)	21.8	21.9	22.0	22.0
# live fetuses (mean)				
Postpartum day 0	13.0	11.6	14.0	15.6
1	12.4	11.4	14.0	15.1
4	10.0	11.0	12.7	15.1
21	8.3	9.1	9.1	10.0
Sex Ratio (M/F)	1.08	1.02	0.88	1.27
Live birth index (%)	92.0	92.0	94.7	96.5
Viability index (%)	76.9	95.1	90.8	97.2
Weaning index (%)	100.0	100.0	97.0	100.0
# with malformations	0	0	0	0

Other Tests on F_1 Offspring: No abnormal findings were reported in the following tests:

a. Sense test
b. Behavior test
c. Emotional activity test
d. Learning ability test
e. Reproductive performance test

22. Kidney Toxicity in Normal Hydropenic Munich-Wistar Rats (Study performed by Dr. Brenner.)

This study has already been reviewed (Pharmacology Reviews, dated 8/29 & 1/18/84).

23. Preclinical AZ Bibliography

Although 150 published reports have been submitted (Vols. 3.41 & 3.42), not all of these are directly on AZ. Most of the pharmacology reports on AZ are based on data submitted in the NDA and have been reviewed above. One pertinent abstract is summarized below.

Penetration of AZ into Cerebrospinal Fluid (CSF) & Brain in Rabbits: Bodem, C.R. et al. Univ. of Missouri Sch. of Med., Columbia, MO. Presented at 23rd ICAAC, Oct. 24-26, 1983.

Methods: Gps of normal & Pseudomonas aeruginosa infected rabbits (with experimental meningitis) were given AZ (300mg/kg) IV over 6 hrs. AZ conc'n was determined in serum, CSF & brain samples.

N 50580 -5

Results:

Group (#)	Serum 4 hour	Serum 6 hour	CSF 4 hour	CSF 6 hour
Normal rabbits (6)	111 ± 18	92 ± 14	1.9 ± 0.3	3.0 ± 0.6
Infected rabbits (6)	84 ± 14	84 ± 17	11.7 ± 1.9	14.6 ± 2.0

Rabbits with meningitis had significantly higher CSF conc'ns of AZ than normal rabbit. Brain conc'ns were also higher in infected rabbits than in normal rabbits, and there was considerable reduction in CSF bacterial titers in the treated, compared to untreated rabbits.

Summary & Evaluation

In 1981, Sykes et al. first reported the isolation of a series of bacterial antibiotics containing a B-lactam ring and suggested the class name of "Monobactam" for this family of monocyclic B-lactams. Imada et al. had already reported isolation of 2 bacterial antibiotics, sulfazecin and isosulfazecin, containing similar monocyclic B-lactam rings. Based on such a novel nucleus (3-aminomonobactamic acid), the applicant has accomplished a total synthesis of the antibiotic aztreonam (SQ 26,776) for which the present Form 5 application has been submitted.

The applicant has performed extensive preclinical studies with aztreonam. There have been 150 published reports submitted. A symposium held in 1981 on aztreonam has also been published (J. Antimicrobial Chemotherapy, Vol. 8, Suppl. E, December, 1981).

This new synthetic B-lactam antimicrobial agent has been shown to be different from the penicillins or cephalosporins in its activity. The compound is relatively inactive against gram-positive and anaerobic gram-negative organisms, but is highly active against the majority of aerobic gram-negative bacteria tested, including Pseudomonas aeruginosa (MIC $<$ 10ug/ml). Aztreonam has been shown to be very similar to other B-lactam antibiotics (cefmetazole, cefotaxime) in terms of environmental effects such as media, pH, inoculum size and presence of human serum. Resistance to its action did develop in organisms under appropriate selective pressure, and at least in E. coli decreased membrane permeability might account for this resistance.

Aztreonam was shown to exhibit a high degree of stability to both plasmid mediated and chromosomally-mediated B-lactamases from gram-negative bacteria. Induction studies with some clinically important isolates showed that aztreonam, similar to cefaperazone, was a poor inducer of B-lactamase in most cases, whereas maximum induction was caused by ampicillin and cefotoxin.

Aztreonam was compatible with other antibiotics such as nafcillin, cloxacillin, erythromycin and vancomycin, and it often acted synergistically with aminoglycosides.

In various animal models, the drug has been shown to be effective in curing or preventing bacterial infections such as urinary tract infection in mice (acute pyelonephritis), lower respiratory tract infection in rats, infection in neutropenic mouse model and surgical wound infection in mice. Various gram-negative organisms were used in these animal models. Aztreonam did not induce antibiotic associated colitis in hamsters when administered as a single 100mg/kg dose IP. In this model, clindamycin, moxalactam and cefaperazone did induce colitis. It appears that anaerobic normal flora have a role in inhibiting C. difficile, since this organism could not be recovered from aztreonam treated animals, although aerobic gram-negative rods were eliminated from the cecum by aztreonam treatment.

Pharmacokinetic studies have been carried out in rats, mice, monkeys, rabbits and dogs. When given parenterally to mice, rats and monkeys, peak serum concentrations occurred in about 10-20 minutes after dosing and 40-50% of the dose was excreted in the urine. In rodents about 13% of the dose was recovered in the bile during a 2-hour period. Aztreonam was well-distributed in tissues with high levels being detected in kidney, liver, genital tract and meninges. Although radioactivity in lymph nodes of female rats after administration of radiolabeled drug was higher than in serum after 2 hours post-dose, no major sex difference in concentration of unchanged aztreonam was seen in rats. In vitro protein binding of aztreonam in human serum was 27% at 2 hours, which increased to 70% at 24 hours. The exact nature of the binding remains unknown. Significant concentrations of the drug were detected in CSF, placentas, fetuses and amniotic fluid. When given to lactating rats, the drug was present in milk and in the pups. After parental administration (IV, IM, SC), the $t_{1/2}$ was about 1 hour in dogs and monkeys between 1 and 6 hours post-dosing. Of the 4 metabolites detected in monkey urine after either IM or IV administration of aztreonam, only the major metabolite has been identified as SQ 26,992, which resulted from opening of the B-lactam ring. None of the metabolites showed any antibacterial activity.

Acutely, the drug was only slightly toxic to mice dosed orally (LD_{50} = 5600mg/kg). The IV LD_{50} of Azactam^R in mice was somewhat lower (1785mg/kg for males; 1710mg/kg for females) than the oral LD_{50}. Aztreonam itself was less toxic acutely than the arginine blend (Azactam^R). Death was preceded by ataxia and convulsions. Histopathological examinations revealed membranous glomerulopathy as the cause of death of one mouse that was examined. Necrosis with calcification of renal tubules at the corticomedullary junction was seen in a surviving rat. Pathological findings in the dead rats included diffuse vacuolar change and individual cell necrosis of the liver, severe tubular degeneration in the kidneys and severe necrosis at the injection site. There was no evidence of potentiation of acute toxicity when the drug was given IV to mice in combination with a number of other antibiotics or probenecid. The IV LD_{50} was similar in healthy mice and those with drug-induced hepatic or renal damage. Also, the neonatal rats were not more susceptible to acute toxicity of the drug, since LD_{50} (IP) was greater than in adults.

Thirty-day subchronic toxicity studies have been performed in rats, with the Na salt of aztreonam given to rats (SC) and dogs (IV), while the arginine blend of aztreonam has been used to perform subacute IV, SC and oral toxicity studies of various durations in rats and dogs.

In the studies with Na salt of aztreonam in rats, dose-related increases in relative liver and kidney weights and in the severity of subcutaneous hemorrhage were rted. Histologically, subacute or chronic inflammation at the injection sites, vacuolation in renal epithelial cells in 1/3 high-dose (2400mg/kg) females, splenomegaly and extramedullary hematopoiesis primarily in the high-dose animals have been reported. No significant microscopic changes were found in the liver. In the dog IV study, collagen degeneration and granulation tissue formation (but no tissue necrosis, edema or excessive inflammation) was observed at the injection sites. In the kidneys of 3/4 dogs of the high-dose (650mg/kg) group, there was patchy chronic inflammation in submucosa below the transitional epithelium of the pelvis, but this was not seen at lower dose levels. Again no liver lesion was seen microscopically.

In the studies performed with the arginine blend of aztreonam, the following significant adverse effects were observed. In the 1-month SC toxicity study in rats, treatment-related changes were seen in the liver, kidney, spleen, formed elements of the blood, and at the injection sites. Splenomegaly associated with extramedullary hematopoiesis, and hematologic changes seen in the high (2400mg/kg) and mid (600mg/kg) dose animals, appears to be secondary to irritation and hemorrhage at the injection sites. Increased liver weights reported in these two groups of animals was not associated with any histopathological changes. Increases in the relative kidney weights of high dose rats were, however, associated with mild vacuolation in the renal tubular epithelial cells. The "no-effect" dose in the rat was found to be 150mg/kg. In the 30-day IV dog study, the only significant treatment-related changes were an increase in the relative liver weights and possible segmental to diffuse disruption of cells from the basement membrane of the kidney. Thus, the kidney and possibly the liver appear to be the target organs for toxicity of this drug. This assessment has been confirmed by a 5-week IV toxicity study in rats performed by NRI Life Sciences in Japan, under contract from the applicant.

In the Japanese study, rats receiving daily doses of 270, 750 or 2000mg/kg of Azactam^{R} had a high incidence of liver and kidney enlargement, and significant enlargement of the cecum, in both the dead and surviving animals. In the high-dose group, there were 17/40 deaths during treatment. Microscopically, moderate centrilobular hypertrophy of the hepatocytes and slight to moderate vacuolation of the tubular epithelium of kidneys were seen in all 750 & 2000mg/kg animals. Additionally, regeneration of the proximal tubular epithelium, moderate hypertrophy of the Bowman's capsule and thickening of basement membrane were seen in 1/3 high-dose males. The liver and kidney vacuoles were not due to increased fat or glycogen deposits. Hyperplasia of the white pulp and hyperactivity of the germinal centers in the spleens of high-dose animals were also reported. Electron microscopic exam of liver and kidneys confirmed these lesions. Myelin-like structures were seen in the glomerular epithelium and in the Kupffer cells. Lysosome-like granules and atypical mitochondrias were seen in the hepatocytes. When the animals were allowed to recover for 35 days, most of these lesions were seen to have regressed. A "no-effect" dose in this study was 100 or possibly 270mg/kg.

Chronic SC studies of 6 months duration have been performed in rats and dogs. In the rat study, severe irritation at the injection sites, decreases in Hb, Hct and erythocytes, marked increase in reticulocytes, extramedullary hematopoiesis and hemosiderosis in the liver, bone marrow leukoplastic and erythroblastic hyperplasia and vacuolation in mucosal epithelial cells of the urinary bladder were seen in the high and mid-dose animals, in addition to the lesions observed in short-term studies. The increased liver weights may be due to increased extramedullary hematopoiesis and proliferation of SER seen in electron microscopy. Sporadic ocular lesions seen in both treated and control animals may be due to some infection, but the applicant has not described any such infection. The increased number of corpora lutea seen in the high-dose females remains unexplained. Peribiliary residual bodies found in the livers could be the unmetabolized drug. The renal tubular lesions included vacuolation and the presence of large heterolysosomes.

Lesions similar to those in rats, but less severe and less frequent in occurrence, were also found in the dog study. Since no ophthalmoscopic examination was done, ocular toxicity, if any, could not be ascertained. In the dog livers, significantly increased glycogen deposition was noted. This may partially explain the increased liver weights. In both species, some of the lesions appeared to be reversing during the recovery period.

Two short-term (2-week) oral toxicity studies have been done in rats and dogs. Except for diarrhea, no significant adverse effects were seen in the dog. In the rat, additionally, dilatation of the cecum was reported. However, no microscopic changes were seen in this organ.

In an independent study, Dr. Barry Brenner of Harvard University had reported earlier that administration of aztreonam/arginine or arginine alone (32mg/kg) for 10 days to hydropenic Munich rats caused morphologic changes in the kidney tubules. His own reevaluation of the results submitted recently indicated that the changes observed were mostly artifactual. Since the applicant's chronic toxicity studies with doses of arginine as high as 150mg/kg did not reveal any kidney toxicity, it is unlikely that Azactam^{R}, when used for short duration according to manufacturer's direction would cause any renal damage. Patients with compromised renal function should, however, be carefully monitored.

Aztreonam was not mutagenic either in the Ames test or in mammalian *in vivo* tests. It is not teratogenic in rat or rabbit. The drug had no significant effect on reproductive performance of the rats. What appeared to be slight embryotoxicity in rabbits at a high SC dose (600mg/kg) most likely was an effect secondary to maternal toxicity (although the investigators attribute it to handling stress).

In a two-generation study in rats, at a dosage level of 750mg/kg, there was a tendency toward decrease in the pup viability index. The "no-effect" dose in these studies appears to be 270mg/kg of aztreonam.

Recommendation

I find this application approvable with the following labeling change. Under "Precautions" (vol. 3.4, page 4010) the first paragraph should read: "Experience in patients with impaired hepatic and renal function is limited; appropriate monitoring of liver and kidney functions in such patients is recommended during therapy."

S.N. Alam, Ph.D.

cc: Orig. NDA 8/8/84
HFN-815
HFN-815/MO
CSO
HFN-220
HFN-815/SNAlam/smc/7/31/84
R/d init.by:JMDavitt
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CHEM. REVIEW

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Attn: N. Lavy, Vice President
Drug Regulatory Affairs

5. SUBMISSIONS REVIEWED

a. ORIGINAL DATED 10/20/83

b. AMENDMENTS DATED 11/30/83

5c. PROVIDING FOR
Manufacturing Controls

6. NAME(S)

a. TRADE
AZACTAM

b. NON-PROPRIETARY
Aztreonam for Injection

c. CHEMICAL
Propanoic Acid, 2-[[1-(2-amino-4-thiazolyl)-2-[(2-methyl-4-oxo-1-sulfo-3-AZETIDINYL)AMINO]-2-oxoethylidene]AMINO]oxy-2-methyl;[2S-[2[, 3B(Z)]]

d. ESTAB

7. STRUCTURAL FORMULA

e. USAN
Aztreonam

f. WHO

8. DOSAGE FORM
Injection: 0.5, 1.0 and 2.0 grams/container

9. ☒ RX ☐ OTC

10. FAMILY OR TYPE OF DRUG
Antibiotic

11. RELATED NDA, IND, MF, FORM 5'S
DMF's 3965, 2390, 957

12. REMARKS
11/30/83 - Firm supplied systems suitability test for HPLC.

A suitable certification monograph will be negotiated with the firm.

13. CONCLUSIONS
Controls are inadequate. See attached 13. Conclusions.

14. DATE REVIEWED 12/9/83

15. REVIEWER Joan M. Eckert

FORM FDH-1742
5/65 RD: init. by RNorton/12/12/83

COPY TO: 1. Original IND HFN-140, HFN-140/CSO,
2. Duplicate IND HFN-430, HFN-535
3. Triplicate IND HFN-140/JMEckert/12/29/83/dv

1. Components & Composition

Aztreonam (100%) (mg/container)	L-Arginine* (mg/container)	Filling Excesses Siliconized Containers 15 ml. vials(%)	100 ml. bottles(%)	Non-Siliconized Containers 15 ml. vials(%)	100 ml. bottles(%)
500	approx. 390	11	3	19	3
1000	approx. 780	7	3	11	3
2000	approx. 1560	6	3	8	3

* The amount of L-Arginine is that quantity needed to obtain a solution pH of about 5 upon constitution with sterile water for injection.

Aztreonam is sparingly soluble (approx. 10 mg/ml) in water of pH 2, and is very soluble (> 1 g/ml) at pH values above 4.

2. Source & Synthesis — Adequate

26771 + 26903 → 26904 → 26776

The synthesis of aztreonam is adequately described on pages 2002-2015 and 8005-8008 of the application.

3. Raw Materials — Adequate

a. The active ingredient and the dosage form will conform to the standards published in the Code of Federal Regulations. These standards have not yet been established.

b. The following materials used in the preparation of aztreonam and L-arginine conform to their respective compendial monographs:

Acetone USP
Anisole FC
L-arginine USP
Dihydrated alcohol USP
Hydrochloric acid NF
Methanol NF
Methylene Chloride NF (Not tested for Water)
Trichloroacetic acid USP
Water for Injection USP

The following materials do not conform to an official compendia but are adequately controlled in the new drug application:

Darco G-60
Dicyclohexylcarbodiimide
Dimethylformamide
Ethanol denatured with light petroleum ether
Ethanol SD 3A
Ethanol SD 23A
Hydroxybenzotriazole Monohydrate
Hyflo
Oxalic acid dihydrate
triethylamine
SQ 26 771
SQ 26 903

L-arginine is obtained from suppliers such as:

Ajinomoto USA Inc., New York, N.Y.
Chemical Dynamics, Inc., South Plainfield, New Jersey
Tonabe USA, San Diego, California.

4. Manufacturing & Processing — Inadequate

Nonsterile aztreonam may be manufactured at Swords Laboratories, Ltd., Dublin, Ireland, Squibb Manufacturing, Inc., Humacao, Peurto Rico, and von Heyden GmbH, Regensburg, Germany. Each of these firms is a wholly-owned subsidiary of E.R. Squibb & Sons, Inc.

Sterile aztreonam will be prepared by Squibb Manufacturing, Inc., Humacao, Peurto Rico.

To prepare sterile aztreonam, crystals of nonsterile aztreonam are dissolved in a hot solution (60°C) of ethanol-water (63:37 v/v). Solution is passed through a 0.2 micron filter, rinsed with the ethanol-water solution and cooled to 5°C with agitation. After filtering and washing with sterile ethanol, the wet cake is combined with hot (50°-55°C) sterile ethanol, cooled and filtered. The wet cake is dried at about 45°C *in vacuo* and the crystals pulverized, if desired.

Sterile Aztreonam is tested by Squibb Manufacturing, Inc., Humacao, Peurto Rico and/or E.R. Squibb & Sons Inc., New Brunswick, N.J.

The sterile L-arginine is prepared by dissolving in warm (60° - 80°C) water for injection. The solution is filtered through a 0.2 micrometer filter into a sterile glass-lined or titanium crystallizer. Acetone is transferred through a filter to the cooled (25° - 30°C) arginine solution. Cool, filter and dry in a suitable vacuum dryer.

The final blend is manufactured, filled and packaged by Squibb Manufacturing, Inc.

The equipment used for preparing the nonsterile aztreonam is described as fabricated from 316 stainless steel, glass-lined steel, or titanium equipped reaction vessels, filters, centrifuges, holding tanks, and dryers. No sizes are given.

The largest batch size for the blended bulk is 500 kg.

No information is provided on their membrane filtration validation tests before and after filtrating.

A description of the controls used to determine if equipment used in sterilizing the materials, that is aztreonam and L-arginine, and the equipment used in the sterile areas is sterile.

5. Laboratory Controls

Firm tests sterile aztreonam for potency, sterility pyrogens, identity, heavy metals, and residue on ignition. Potency is done by HPLC. Sterile aztreonam is not tested for moisture or pH.

Aztreonam for injection is tested for potency, sterility, pyrogens, safety, moisture, and pH.

Cerification monographs will be negotiated with the firm.

The HPLC method used for aztreonam can also be used to determine the content of L-arginine by adding L-arginine to the aztreonam working standard solution.

The HPLC method is capable of separating impurities and degradation products from aztreonam, as listed:

SQ 26,992 - major degradation product of aztreonam;
SQ 26,776 (Aztreonam);
I-1- an unidentified impurity;
SQ 28,429 - the "E" isomer of aztreonam;
SQ 27,412 - the ethyl ester of aztreonam.

The potency of aztreonam is not less than 900 micrograms per milligram of aztreonam.

In the synthesis of SQ 26,776 (Z isomer), SQ 26,903 and SQ 26,771 are reacted to form 26,904 which is the penultimate compound (the benzhydryl ester of 26,776). The structures of both 26,903 and 26,771 have been confirmed by single crystal X-ray diffraction.

To yield aztreonam free of the E-isomer, the benzhydryl ester group is removed from SQ 26, 904.

The proof of structure of aztreonam has been confirmed by mass spectrometry.

There are three forms of aztreonam, α, β, ___ at various stages during synthesis. These three forms can be distinquished from one another by DSC and IR. The final stage produces β aztreonam which is virtually free of other polymorphs. The identity test performed by the firm is an IR test which distinquishes the α, β + forms. The HPLC test which is the test for content is capable of separating the E-isomer from aztreonam.

6. Stability

Although aztreonam will be synthesized at three locations, Dublin, Ireland, Humacao, P.R., and Regensburg, Germany, only data from two batches produced in Germany were filed.

Two lots of sterile aztreonam, MB00400 and MB00500, were held for 12 months at 5°C, 33°C, 40°C and 40°C/75% RH. The testing was done at 3, 6, 9, and 12 months except for the material held at 40°C/75% RH which was tested at 6 and 12 months. There was no room temperature storage.

There was no significant loss in potency.

No expiration period has been requested for the bulk product.

No description of packaging of this material was supplied.

No storage conditions have been given for the bulk.

An expiration period of 2 years is requested for the blend.

Six lots of blended material were stored at room temperature (22°C), 33°C, 40°C and 50°C, upright and inverted for the following periods:

** 0.5 gram (MNB-863-0/RB3)	15 ml	Upright	7 months
" " " " "	"	Inverted	7 months
** 0.5 gram (MNB-863-0/RB11)	15 ml	Upright	7 months
" " " " "	"	Inverted	7 months
0.5 gram (MNB-863-0/C56)	15 ml	Upright	5 months
" " " " "	"	Inverted	5 months
0.5 gram (MNB-863-0/C57)	15 ml	Upright	5 months
" " " " "	"	Inverted	5 months
** 1.0 gram (MNB-864-0/C33)	15 ml	Upright	6 months
" " " " "	"	Inverted	6 months

** 1.0 gram (MNB-864-0/C33)	100 ml	Upright Inverted	6 months 6 months
** 1.0 gram (MNB-864-0/C35)	15 ml	Upright Inverted	6 months 6 months
** 1.0 gram (MNB-864-0/C35)	100 ml	Upright Inverted	6 months 6 months
** 2.0 grams (MNB-865-0/C33)	15 ml	Upright Inverted	6 months 6 months
** 2.0 grams (MNB-865-0/C33)	100 ml	Upright Inverted	6 months 6 months
** 2.0 grams (MNB-865-0/C35)	15 ml	Upright Inverted	6 months 6 months
** 2.0 grams (MND-865-0/C35)	100 ml	Upright Inverted	6 months 6 months

* PH performed at initial time only.

** Moisture performed at initial time and 6 months rather than initial time only.

A two year expiry is requested for the blended product for IM and IV use.

Solutions of the aztreonam blend were prepared and held on stability as follows:

I.M. - Glass containers

Twenty-five percent (W/V) solutions containing 250 mg/ml were prepared with 6 different diluents. The solutions were prepared by dissolving 77.8 grams of azactam (539.9 mg of aztreeonam/gram of aztreonam/L-arginine blend) in sufficient diluent to yield 158 ml of so ution.

Type I glass, 15-ml vials were filled with 4.2 milliliters of solution, (4.2 milliliters was used because it is the average volume obtained after reconstituting with 3 milliliters of diluent for IM use.) The stoppers used were 20 mm, West 1869 Gray Butyl rubber stoppers. There is no mention of an aluminum seal.

The solutions were stored at the following temperatures and tested initially and on the days specified:

40°C - Days 2, 3, 7
R.T. - Days 3, 7, 10, 14
5°C - Days 7, 14, 21, 28

The diluents were:

1. 0.9% sodium chloride injection, USP
2. Bacteriostatic sodium chloride injection, USP with benzyl alcohol.
3. " " " " " " " " , USP, paraben preserved.
4. Sterile water for injection, USP.
5. Bacteriostatic Water for Injection USP with benzyl alcohol.
6. Bacteriostatic Water for Injection, USP, with Parabens.

I.V. - Glass containers

1% and 2% solutions were prepared with 26 different diluents. The solutions were prepared by dissolving 46.68 grams of the blend in sufficient diluent to yield 2429 ml. of solution for the 1% solution and 93.36 grams of the blend is dissolved in sufficient diluent to make 2458 ml. of solution. For the 1% solution, bottles were filled with 101.2 ml. of solution and for the 2% solution, bottles were filled with 102.4 ml. of solution. The bottles used were Squibb code A 4104, 100 ml., Type I glass bottles. The stoppers were Squibb code H 5174, 28 mm., West 1869 Gray butyl rubber stoppers.

The solutions were stored at the following temperatures and tested initially and on the days specified.

	Diluents IVA-IVQ	Diluents IV-R-IV-Z
40°C	Days 2, 3, 7	Days 2, 7
RT	Days 3, 7, 10, 14	Days 2, 7, 15
5°C	Days 7, 14, 21, 28	Days 7, 16, 28

The diluents are listed on pages 8103 - 8109.

The firm states that these data support the storage conditions recommended in the labeling. Since the labeling was not included in this portion of the application, the storage times are unknown to us.

Based on the data filed for the IM and IV, it appears that the solutions retained 90% of initial potency for the following periods:

IM - 40°C None all assays are below 80% at 2 days
R.T. 3 days. At 7 days (the next test time after 3 days), all assays were below 80%
5°C 14 days. At 21 days (the next test time after 7 days), all assays were either below 90% or borderline. None were over 92.6%.

1% - IV Diluents a - Q
40°C - 3 days except for 10% dextrose injection USP (IV-F), which should be 2 days.
R.T. - 14 days except for: 10% dextrose injection USP (IV-F)
Isolyte E with 5% dextrose (IV-K)
5% mannitol Injection USP (IV-Q)

IV-F 7 days
IV-K 7 days
IV-Q 10 days
5°C 28 days

1% IV Diluents R - Z
40°C - 2 days except IV-V & IV-W-None
RT - 7 days
5°C - 28 days

2% IV Diluents A - Q
40°C - 3 days except for 10% travert injection (IV-P), which should be 2 days.
R.T. 14 days except:
5% dextrose injection (IV-E) - 10 days
10% " " " (IV-F) - 7 days
5% " " and 0.9% sodium chloride injection USP (IV-G) - 10 days
Isolyte E with 5% dextrose (IV-K) - 7 days
Isolyte M with 5% dextrose (IV-L) - 10 days
10% Travert Injection (IV-P) - 7 days
5% Mannitol injection (IV-Q) - 10 days

5°C - 28 days

2% IV Diluents R-Z
40°C - 2 days; except IV-V & IV-W-None
RT - 7 days
5°C - 28 days

5% sodium bicarbonate is unsuitable as a diluent at any temperature.

Pre-Filled Flexible Plastic Containers

Travenol's Via-Flex containers containing 100 milliliters of each diluent tested were used.

A 1% and a 2% solution was prepared using the same bulk as the stability study with glass containers, MND-860-H/C06-539.9 mg/gram of blend.

1% solution - 1,945 mg of the blend is dissolved in and diluted to 4.2 milliliters of the diluent being tested. The resultant solution is added to the flexible container containing 100 milliliters of the same diluent.

2% solution - Same as 1% except 3,890 mg of the blend is dissolved in and diluted to 8.4 milliliters with the diluent to be tested.

The solutions were stored at the following temperatures and tested initially and on the days specified.

40°C - Days 2, 3, 7
R.T. - Days 3, 7, 10, 14
5°C - Days 7, 14, 21, 28

The diluents were: 0.9% Sodium Chloride Injection, USP and 5% Dextrose Injection, USP.

Stored samples remained stable for the following expiration periods:

0.9% Sodium Chloride Injection USP

1% - 40°C - 3 days
R.T. - 14 days
5°C - 28 days

2% - 40°C - 2 days
R.T. - 14 days
5°C - 28 days

5% Dextrose Injection USP

1% - 40°C - 3 days
R.T. - 14 days
5°C - 28 days

2% - 40°C - 2 days
R.T. - 10 days
5°C - 28 days

The firm has not specified any expiration period. However this data supports an expiration period of two days when stored at controlled room temperature and seven days when stored under refrigeration.

Empty Flexible Plastic Containers

Travenol's Via - Flex containers were used.

The 1% and 2% solutions were prepared and the containers filled identical to the glass bottles.

The solutions were stored at the following temperatures and tested initially and the days specified:

40°C - Days 2, 3, 7
R.T. - Days 3, 7, 10, 14
5°C - Days 7, 14, 21, 28

The diluents tested were:

0.9% Sodium Chloride Injection, USP
Ringers Injection, USP
Lactated Ringers Injection, USP
5% Dextrose Injection, USP
5% Dextrose and 0.9% Sodium Chloride Injection, USP
5% Dextrose and 0.45% Sodium Chloride Injection, USP
Normosol - M and 5% Dextrose.

The firm has not supplied any expiration period.

Data demonstrated that solutions retained at least 90% of potency or greater for the following periods:

	1%			2%		
	40°C	R.T.	5°C	40°C	R.T.	5°C
0.9% Sodium Chloride Injection USP	3 days	14 days	28 days	3 days	14 days	28 days
Ringer's Injection USP	3 days	14 days	28 days	3 days	14 days	28 days
Lactated Ringer's Injection USP	7 days*	14 days	28 days	3 days	14 days	28 days
5% Dextrose Injection USP	3 days	14 days	28 days	2 days	7 days	28 days
5% Dextrose Injection and 0.9% Sodium Chloride Injection USP	2 days**	14 days	28 days	2 days**	7 days***	28 days
5% Dextrose Injection and 45% Sodium Chloride Injection USP	2 days**	16 days	28 days	2 days**	7 days***	28 days
Normosol-M and 5% Dextrose	2 days**	16 days	28 days	2 days**	7 days***	28 days

* These results are out of line with all other data

** Data not supplied for 3 days only 2 days

*** Data not supplied for 10 days

Data supports on expiration period for the above infusion solutions of 2 days at room temperature and 7 days under refrigeration.

7. Control Numbers — Adequate

8. Containers & Closures — Inadequate

a. Storage containers for nonsterile aztreonam - No description given.

b. Storage containers for sterile aztreonam - No description given. The sterilization procedure is described as follows: The containers are cleaned with hot water and steam and rinsed with water for injection. They are sterilized by steam under pressure of not less than 15 PSIG at 121 C for at least 30 minutes. Sterilization validation procedures should be described.

c. Storage containers for the blended material-

No description given.

d. Vials - Type I glass vials of 15 ml. or 100 ml. capacity. Vials will be supplied by Wheaton Tubing Products and Wheaton Glass Company, both of Millville, New Jersey, or the Kimble Division of Owens-Illinois of Chicago Heights, Illinois. The closures are made from West Company's 1869 Gray formulation. The closure is held in place by an aluminum crimp seal purchased from West Company, Phoenixville, Penn.

The closures are sterilized by steam at a minimum of 121 C for not less than 30 minutes. No pressure is given.

The glass containers are dry-heat sterilized by heating at a minimum of 250 C for at least 30 minutes.

The containers and closures may be siliconized according to Section 6.A.2 and 6.C.3 respectively of Master File 893.

9. Environmental Impact Statement — Adequate

A statement of exemption was filed.

10. Labeling — Inadequate

No labeling was filed.

13. Conclusions

Firm should file the following information:

1. The sizes of the equipment used for preparing the nonsterile aztreonam, such as reaction vessels, filters, centrifuges, holding tanks and dryers.

2. The largest intended batch size for the finished vials.

3. Information concerning membrane filtration validation tests before and after filtering.

4. A description of the controls used to determine, the sterility of equipment used in sterilizing the materials, aztreonam and L-arginine, and the equipment used in the sterile areas.

5. A description of the packaging for the two lots of aztreonam bulk, MB00400 and MB00500, held for stability testing.

6. The intended storage conditions and suggested expiration period for the bulk (not blended).

7. The intended expiration periods and storage conditions recommended for the constituted solutions for each diluent.

8. Validation procedures for the sterilization of the vials and bottles for the finished product.

9. A description of storage containers for the dry blended material.

MICROBIOLOGY

Aztreonam (SQ 26,776) is intended for use in patients for the parenteral treatment of bacterial infections caused by aerobic gram-negative organisms.

SQ 26,776 is a zwitterion. It contains both a positive and a negative charge located on different parts of the molecule. The dipotassium salt of SQ 26,776 is designated SQ 26,726. In aqueous solution the salt form, SQ 26,726, ionizes to give SQ 26,776. SQ 26,726 and SQ 26,776 exhibit the same antibacterial spectrum of activity.

The following preclinical studies were done using SQ 26,726 not SQ 26,776:

IN-VITRO MICROBIOLOGY

SQ 26,726 was compared with some cephalosporins penicillins and gentamicin against gram-positive and gram-negative aerobes and anaerobic gram-negative organisms (See tables 1, 7, 11 and 12).

SQ 26,726 is active against a variety of Gram-negative organisms. It has little or no activity against aerobic gram-positive organisms or gram-negative anaerobic organisms.

EFFECTS OF MEDIA/PH ON ACTIVITY

Five media at three pH levels were tested using an overnight culture of E. coli SC 8294 diluted to about 10^5 CFU per ml. Serial twofold dilutions of SQ 26,726 were compared to cefotaxime and gentamicin (TABLE 2).

SQ 26,726 does not appear to be effected by differences in pH or media. However, in the case of Mueller Hinton Broth, there is a twofold increase in activity in the MIC's between pH 6 and pH 7 and a fourfold increase in activity in the MBC between pH 6 and pH 7.

There is a fourfold increase in the MIC for nutrient broth between pH 6 and pH 7. The MBC's increase threefold. SQ 26,726 is most active at pH 7 in Mueller Hinton Broth and Nutrient Broth.

SQ 26,726 was diluted in McIlvaines buffer (pH 3.0, 5.0, and 7.0) and in a boric acid-borax buffer system (pH 7.5, 8.0, and 9.0). The samples were analyzed by HPLC over a 24 hour period at 0, 0.5, 1, 2, 4, 6, and 24 hours. The only significant loss in potency was at pH 9.0 at 24 hours (78%). TABLE 3.

INOCULUM SIZE

Overnight cultures of 10^3, 10^5, and 10^7, CFU's per ml of the test strains were diluted with K-10 broth (0.15% beef extract, 0.3% yeast extract, 0.6% peptone, and 0.1% dextrose).

The results show that inoculum size does have an adverse effect on SQ 26,726, (TABLE 4).

HUMAN SERUM

MIC's and MBC's for SQ 26,726, cefotaxime, and gentamicin were determined against seven gram-negative organisms in antibiotic assay broth containing 50% human serum.

There was no change in the antibacterial activity for SQ 26,726, (TABLE 8).

In a second study, a solution containing 1 mg/ml of SQ 26,726 was prepared in human, mouse and rat serum and analyzed by HPLC for a 6-hour period.

After 6 hours, 86% remained in the human serum, 50% in the mouse serum and 40% in the rat serum.

INTERACTION WITH OTHER ANTIMICROBIAL AGENTS

SQ 26,726 was combined with cephradine, cefoxitin, gentamicin, clindamycin and metronidazole and evaluated for activity against a variety of gram-positive and gram-negative aerobic and anaerobic organisms.

The only combination that demonstrated true synergy was with metronidazole against certain strains of Bacteroides fragilis. Firm states that "for all other combinations, the results were additive or indifferent. In no case was antagonism observed." They reference to an Internal report, but no data are given to demonstrate which combinations were additive. The Internal Report is in missing volume 1.1.

RESISTANCE DEVELOPMENT

E. coli, S. marcesens and P. aeruginosa developed resistance after several passages in broth for 6-10 consecutive days. Data are not given, just a reference to the internal report.

IN VITRO ACTIVITY

ACTIVE

	Standard Strains		Recent Clinical isolates	
	MIC 50	MIC 90	MIC 50	MIC 90
E. coli	≤0.1	0.2	0.1	0.3
Klebsiella	≤0.1	0.3	0.1	2.7
Klebsiella Enterobacter group	0.2	0.4	-	-
Serratia	0.2	0.4	0.4	1.5
Citrobacter	0.2	0.6		
Indole + (Proteus vulgaris)	≤0.1	0.8		
Salmonella	0.2	0.3		
H. influenzae	≤0.1	0.1		
Ampicillin resistant H. influenzae	≤0.1	≤0.1		
PPN and NPPN gonorrhea	≤0.1	0.2		

LESS ACTIVE

Enterobacter	≤0.1	31.3	0.4	23.1
Shigella	0.2	10.7		
Pseudomonas*	5.5	18.8	5.5	16.7

VERY ACTIVE

Indole negative Proteus	≤0.1	≤0.1	≤0.05	≤0.05
Providencia	≤0.1	≤0.1	≤0.05	≤0.05

*SQ 26,726 appears to be active against some gentamicin-resistant Pseudomonas aeruginosa strains. Out of 11 strains tested, 6 had MIC's of 3.1 ug/ml, 4 with 6.3 ug/ml, and one 12.5 ug/ml. Since no clinical studies are available, there is no way to correlate MIC's with clinical cures.

The most active drug tested against these strains of Pseudomonas was ceftazidime.

The guidelines for susceptibility testing supplied to the investigators for protocols 18,554-10A and 18,554-11A are. Susceptible $\leq$ 6.3 µg/ml; Resistant - 25 µg/ml; and Intermediate 12.5 - 25 µg/ml.

SUSCEPTIBILITY TESTING

Firm filed Regression lines with a 2, 5, 10, and 30 ug disc for various gram-organisms.

According to the criteria sent to the investigator for protocols 10 and 11, October 27, 1982, all the organisms would be regarded as susceptible.

The 30 ug disc may not be the most appropriate one in that the zone diameters are somewnat large. However, until some correlation between clinical and in-vitro data can be establisned, comment will be withheld.

MODE OF ACTION

SQ 26,726 binds to specific enterobacterial membrane proteins which are involved in cell wall syntnesis. These proteins also bind penicillin and are called penicillin binding proteins or PBP's.

Table 1

Activity of SQ 26,726 and Selected β-Lactam Antibiotics
vs. Primary Screen Microorganisms

*MIC (μg/ml)

Organism	SC #	SQ 26,726	Cefuroxime	Ceftazidime	Cephamandole	Cefuzolin	Cefotaxime	Cefoperazone	Moxalactam	GR20,263	Piperacillin	Azlocillin
Staph. aureus	1276	>100	0.8	0.4	<0.05	1.6	0.8	0.8	3.1	12.5	0.4	0.2
Staph. aureus	2395	>100	0.8	0.4	0.2	1.6	0.8	0.8	3.1	12.5	0.4	0.4
Staph. aureus	2400	>100	0.8	0.8	0.4	3.1	0.8	1.6	3.1	12.5	3.1	3.1
Staph. aureus	10,165	>100	3.1	3.1	1.6	12.5	3.1	12.5	25	50	100	25
Staph. epidermidis	9611	>100	3.1	>100	25	>100	0.2	12.5	>100	6.3	0.8	0.8
Staph. saprophyticus	9767	25	<0.05	0.4	<0.05	0.8	<0.05	<0.05	0.8	0.1	<0.05	<0.05
Micrococcus luteus	2495	12.5	0.1	0.4	<0.05	0.8	<0.05	<0.05	0.8	0.4	<0.05	<0.05
E. coli	8294	0.4	6.3	1.6	1.6	12.5	<0.05	0.8	0.8	0.4	3.1	12.5
E. coli	10,157	0.1	0.2	0.8	0.4	3.1	<0.05	<0.05	0.2	0.1	<0.05	0.2
E. coli	10,896	0.2	3.1	0.8	0.8	6.3	<0.05	<0.05	0.1	0.1	0.8	12.5
E. coli	10,909	<0.05	3.1	0.4	0.8	6.3	<0.05	<0.05	0.1	0.1	0.4	1.6
K. pneumoniae	10,430	0.4	6.3	3.1	1.6	12.5	<0.05	0.8	0.8	0.4	12.5	25
K. pneumoniae	9527	<0.05	0.4	0.8	0.4	3.1	<0.05	<0.05	0.2	0.1	1.6	12.5
Proteus mirabilis	8855	<0.05	0.8	1.6	0.8	3.1	<0.05	0.8	0.2	<0.05	0.4	3.1
Proteus vulgaris	8479	<0.05	<0.05	0.4	0.2	1.6	<0.05	0.1	<0.05	<0.05	0.1	0.8
Proteus vulgaris	9416	<0.05	0.4	1.6	0.8	3.1	<0.05	<0.05	0.2	<0.05	0.05	<0.05
Salmonella	1195	<0.05	1.6	0.4	0.4	>100	<0.05	0.4	0.1	0.1	0.8	3.1
Shig. sonnei	6449	0.2	3.1	0.8	0.8	6.3	<0.05	0.4	0.2	0.2	1.6	12.5
Ent. cloacae	8236	0.2	6.3	100	12.5	>100	0.1	0.8	0.4	0.4	1.6	12.5
Ent. aerogenes	10,078	0.4	12.5	25	12.5	100	0.1	0.8	0.8	0.8	6.3	25
Citrob. freundii	9518	0.2	6.3	12.5	25	50	0.4	0.4	1.4	1.6	3.1	12.5
Serr. marcescens	9783	0.2	100	12.5	100	25	0.2	3.1	0.4	0.2	1.6	12.5
Ps. aeruginosa	9545	0.4	12.5	50	100	50	0.4	0.8	1.6	1.6	0.4	0.4
Ps. aeruginosa	8429	3.1	>100	>100	>100	>100	12.5	3.1	6.3	1.6	3.1	3.1
Acinetobacter anitratus	8333	50	>100	>100	>100	>100	50	>100	50	12.5	50	50

*Determined by Agar Dilution 10^4 CFU.

Table 2 Effect of Media and pH on Antibacterial Activity

Media	pH	SQ 26,726 (µg/ml) MIC	SQ 26,726 (µg/ml) MBC	Cefotaxime (µg/ml) MIC	Cefotaxime (µg/ml) MBC	Gentamicin (µg/ml) MIC	Gentamicin (µg/ml) MBC
K-10[1]	6	0.31	1.25	0.08	0.31	3.1	3.1
BHI[2]	6	0.63	0.63	0.16	0.16	12.5	25
MI[3]	6	0.63	2.5	0.16	0.31	1.6	3.1
TSB[4]	6	0.31	0.63	0.08	0.16	12.5	25
NB[5]	6	0.63	2.5	0.16	0.31	0.31	0.63
K-10	7	0.31	0.63	0.16	0.31	0.4	0.8
BHI	7	0.31	0.63	0.16	0.16	3.1	3.1
MI	7	0.16	0.31	0.08	0.31	1.6	1.6
TSB	7	0.31	0.31	0.16	0.16	12.5	12.5
NB	7	0.08	0.63	0.16	0.31	0.08	0.16
K-10	8	0.31	0.63	0.16	0.63	<0.2	<0.2
BHI	8	0.31	0.63	0.08	0.16	1.6	1.6
MI	8	0.16	1.25	0.08	2.5	1.6	1.6
TSB	8	0.16	0.31	0.16	0.31	1.6	3.1
NB	8	0.16	1.25	0.08	0.16	0.04	0.04

[1]K-10: Beef Extract-Yeast Extract-Peptone-Dextrose Broth

[2]BHI: Brain Heart Infusion Broth

[3]MI: Mueller-Hinton Broth

[4]TSB: Trypticase Soy Broth

[5]NB: Nutrient Broth

Table 3 pH Stability of SQ 25,725

Time (Hours)	% SQ 25,725 Remaining					
	pH = 3.0[1]	pH = 5.0[2]	pH = 7.0[3]	pH = 7.5[4]	pH = 8.0[4]	pH = 9.0[4]
0	100	100	100	100	100	100
0.5	98	99	104	104	107	104
1.0	98	98	102	110	109	107
2.0	100	100	100	105	103	102
4.0	90	100	103	111	101	96
6.0	97	99	100	105	103	96
24.0	100	102	103	105	99	78

[1]The initial concentration of SQ 25,725 in pH = 3.0, buffer was 0.99 mg/ml.
[2]The initial concentration of SQ 25,725 in pH = 5.0, buffer was 1.02 mg/ml.
[3]The initial concentration of SQ 25,725 in pH = 7.0, buffer was 1.00 mg/ml.
[4]The initial concentration of SQ 25,725 in pH = 7.5, 8.0 and 9.0 buffer was 0.30 mg/ml.

Table 4 Effect of Inoculum* Size on Antibacterial Activity

Organism	Inoculum	SQ 26,726 MIC	SQ 26,726 MBC	Cefotaxime MIC	Cefotaxime MBC	Gentamicin MIC	Gentamicin MBC
E. coli SC 8294	10^3	<0.2	0.4	<0.2	<0.2	0.6	0.8
	10^5	0.2	0.4	<0.2	<0.2	0.8	1.6
	10^7	0.2	0.4	<0.2	3.1	3.1	6.3
E. coli SC 10,404	10^3	<0.2	<0.2	<0.2	<0.2	<0.2	0.4
	10^5	0.3	0.4	<0.2	<0.2	0.4	0.8
	10^7	0.3	0.8	<0.2	<0.2	1.2	1.6
Shig. sonnei SC 8274	10^3	<0.2	<0.2	<0.2	<0.2	1.2	1.6
	10^5	0.2	0.8	<0.2	<0.2	1.2	1.6
	10^7	<6.3	6.3	<0.2	<0.2	2.4	3.1
K. aerogenes SC 10,440	10^3	0.3	0.4	<0.2	<0.2	0.6	0.8
	10^5	0.4	0.8	0.6	0.8	0.6	0.8
	10^7	<25	25	0.6	0.8	1.6	3.1
Ent. cloacae SC 8236	10^3	<0.2	<0.2	<0.2	<0.2	0.6	0.8
	10^5	0.3	0.4	0.3	0.8	1.2	1.6
	10^7	<50	50	6.3	50	3.1	12.5
Proteus, Indole negative SC 9813	10^3	<0.2	0.4	0.8	3.1	2.4	6.3
	10^5	0.4	12.5	6.3	25	3.1	6.3
	10^7	<12.5	12.5	12.5	>50	12.5	25
Ser. marcescens SC 9873	10^3	<0.2	<0.2	<0.2	0.8	1.2	1.6
	10^5	0.3	0.4	0.6	12.5	1.6	3.1
	10^7	<25	25	3.1	>50	9.4	12.5
Ps. aeruginosa SC 9545	10^3	0.4	0.8	0.4	0.8	0.4	1.6
	10^5	0.8	1.6	0.8	3.1	0.8	1.6
	10^7	>50	>50	>50	>50	1.6	3.1
Ps. aeruginosa SC 8329	10^3	4.7	12.5	9.4	12.5	1.2	1.6
	10^5	12.5	25	37.5	50	1.2	3.1
	10^7	>50	>50	>50	>50	1.6	6.3

*Colony forming units

Table 7 Activity Against β-Lactamase Producing Organisms

MIC (µg/ml): Agar dilution		Minimum Inhibitory Concentration (µg/ml)											
Organism	SC #	Compound / Inoculum	Amoxicillin 10^4	Amoxicillin 10^6	Cephaloridine 10^4	Cephaloridine 10^6	Cephamandole 10^4	Cephamandole 10^6	Cefuroxime 10^4	Cefuroxime 10^6	Cefoxitin 10^4	Cefoxitin 10^6	Cefmetazole 10^4	Cefmetazole 10^6
E. coli TEM+	10,404		≥100	≥100	6.3	100	12.5	≥100	3.1	6.3	3.1	6.3	0.8	1.6
E. coli TEM-	10,439		1.6	1.6	1.6	3.1	0.8	1.6	3.1	6.3	6.3	6.3	0.8	1.6
E. coli RGN238	10,854		25	100	3.1	3.1	1.6	6.3	6.3	25	6.3	12.5	1.6	1.6
C. freundii	10,204		≥100	≥100	100	≥100	1.6	≥100	1.6	100	≥100	≥100	50	100
Shig. sonnei	10,944		50	100			6.3	50	50	50	50	50	12.5	25
Sal. typhimurium	10,943		0.4	1.6			1.6	1.6	6.3	6.3	3.1	6.3	0.8	0.8
Ent. cloacae P99+	10,435		≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100
Ent. cloacae P99-	10,441		1.6	1.6	3.1	3.1	6.3	6.3	12.5	25	12.5	12.5	6.3	6.3
Ent. cloacae	8415		25	≥100			3.1	12.5	6.3	100	≥100	≥100	≥100	≥100
Ent. cloacae	9965		-	-			3.1	50	3.1	25	≥100	≥100	≥100	≥100
K. aerogenes K1+	10,436		≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100	1.6	3.1	0.8	0.8
K. aerogenes K1-	10,440		0.8	1.6	1.6	3.1	0.8	0.8	1.6	3.1	3.1	3.1	0.8	0.8
K. pneumo.	8340		50	100			0.8	0.8	1.6	1.6	1.6	3.1	0.4	0.8
K. pneumo.	11,066		≥100	≥100			12.5	≥100	3.1	6.3	6.3	12.5	1.6	3.1
Prot. rettgeri	8217		50	100	≥100	≥100	6.3	100	3.1	100	25	50	25	100
Prot. rettgeri	11,104		-	-			25	≥100	12.5	100	25	50	25	50
Prot. vulgaris	10,950		100	≥100	≥100	≥100	100	≥100	≥100	≥100	3.1	6.3	1.6	3.1
Prot. vulgaris	10,9510		≥100	≥100			100	≥100	≥100	≥100	3.1	6.3	0.8	3.1
Prot. mirabilis	9574		0.2	0.2			1.6	1.6	3.1	25	3.1	12.5	1.6	12.5
Ps. aeruginosa	9545		3.1	100	≥100	≥100	50	100	6.3	25	6.3	25	6.3	25
Ps. aeruginosa	8329		50	≥100			≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100
Ps. aeruginosa	9546		-	-			≥100	≥100	≥100	≥100	≥100	≥100	≥100	≥100
Ser. marcescens	9782		25	50	≥100	≥100	100	≥100	≥100	≥100	12.5	25	6.3	12.5
Ser. marcescens	8247		-	-			≥100	≥100	≥100	≥100	50	≥100	25	100
Acin. calco.	8133		6.3	25	≥100	≥100	≥100	≥100	50	≥100	100	≥100	≥100	≥100

Table 7 (cont'd). Activity Against β-Lactamase Producing Organisms

MIC (μg/ml): Agar dilution		Minimum Inhibitory Concentration (μg/ml)											
		Compound	Cefotaxime		Cefoperazone		Ceftazidime		Moxalactam		SQ 26,726		
Organism	SC #	Inoculum	10^4	10^6	10^4	10^6	10^4	10^6	10^4	10^6	10^4	10^6	10^4	10^6
E. coli TEM+	10,404		≤0.05	0.1	3.1	≥100	0.4	0.4	0.2	0.2	≤0.05	0.2		
E. coli TEM-	10,439		≤0.05	0.1	0.2	0.4	0.2	0.4	0.4	0.4	≤0.05	0.2		
E. coli RGN238	10,854		0.2	1.6	0.8	3.1	0.4	0.4	0.4	0.4	0.2	0.4		
C. freundii	10,204		0.2	6.3	0.2	12.5	0.2	25	3.1	6.3	0.1	25		
Shig. sonnei	10,944		1.6	6.3	0.8	50	1.6	6.3	0.4	0.8	6.3	6.3		
Sal. typhimurium	10,943		0.1	0.2	0.4	0.4	0.4	0.4	0.2	0.2	0.1	0.2		
Ent. cloacae P99+	10,435		50	>100	50	>100	50	100	25	50	12.5	50		
Ent. cloacae P99-	10,441		0.2	0.2	0.2	0.4	0.2	0.4	0.4	0.8	≤0.05	0.2		
Ent. cloacae	8415		0.2	0.8	0.2	0.8	0.4	0.4	0.4	6.3	≤0.05	0.2		
Ent. cloacae	9965		0.1	0.8	0.4	0.8	0.2	3.1	0.4	0.8	≤0.05	0.2		
K. aerogenes K1+	10,436		1.6	100	>100	>100	0.4	1.6	0.8	3.1	50	>100		
K. aerogenes K1-	10,440		≤0.05	≤0.05	0.1	0.2	0.1	0.1	0.8	3.1	≤0.05	0.1		
K. pneum.	8340		≤0.05	≤0.05	0.1	0.4	0.1	0.2	0.4	0.8	≤0.05	0.1		
K. pneum.	11,066		0.1	0.2	12.5	>100	1.6	6.3	1.6	25	0.4	0.8		
Prot. rettgeri	8217		0.1	0.8	1.6	12.5	≤0.05	0.4	≤0.05	3.1	≤0.05	≤0.05		
Prot. rettgeri	11,104		0.4	12.5	25	>100	0.4	3.1	0.2	6.3	≤0.05	0.1		
Prot. vulgaris	10,950		≤0.05	>100	0.4	1.6	≤0.05	0.1	0.4	3.1	≤0.05	≤0.05		
Prot. vulgaris	10,9510		≤0.05	100	3.1	>100	≤0.05	0.2	0.2	3.1	≤0.05	0.2		
Prot. mirabilis	9574		≤0.05	0.8	0.8	1.6	≤0.05	≤0.05	0.2	3.1	≤0.05	≤0.05		
Ps. aeruginosa	9545		0.8	3.1	0.4	0.8	0.8	0.8	1.6	50	0.4	0.4		
Ps. aeruginosa	8329		12.5	>100	3.1	25	0.8	1.6	12.5	>100	6.3	12.5		
Ps. aeruginosa	9546		12.5	25	3.1	12.5	1.6	3.1	3.1	12.5	6.3	12.5		
Ser. marcescens	9782		0.2	25	1.6	12.5	0.2	0.4	0.4	1.6	0.1	0.2		
Ser. marcescens	8247		12.5	>100	25	>100	0.8	3.1	6.3	12.5	1.6	12.5		
Acin. calco.	83		25	100	50	>100	6.3	12.5	100	>100	25	50		

TAB 8

tibacterial Activity in the Presence
- of 50% Human Serum

50% Human Serum	SQ 26,726 (µg/ml) MIC	SQ 26,726 (µg/ml) MBC	Cefotaxime (µg/ml) MIC	Cefotaxime (µg/ml) MBC	Gentamicin (µg/ml) MIC	Gentamicin (µg/ml) MBC
-	0.1	0.2	0.1	0.1	0.4	0.8
+	0.1	0.1	<0.05	<0.05	0.8	1.6
-	<0.05	<0.05	<0.05	<0.05	0.4	0.4
+	<0.05	<0.05	<0.05	<0.05	0.2	0.4
-	<0.05	<0.05	<0.05	<0.05	0.4	0.4
+	<0.05	<0.05	<0.05	<0.05	0.4	0.4
-	0.1	0.1	<0.05	<0.05	3.1	3.1
	0.1	0.2	<0.05	<0.05	3.1	3.1
-	<0.05	0.2	0.2	0.4	0.8	0.8
+	0.2	0.2	1.6	1.6	0.4	0.4
-	<0.05	<0.05	<0.05	<0.05	0.4	0.4
+	<0.05	<0.05	0.1	0.1	0.4	0.4
-	<0.05	<0.05	<0.05	0.1	1.6	1.6
+	<0.05	0.1	0.1	0.1	0.8	1.6
-	0.4	0.8	0.8	0.8	0.4	0.4
+	0.4	0.8	0.8	0.8	3.1	12.5
-	1.6	1.6	6.3	6.3	0.4	0.8
+	0.8	1.6	3.1	6.3	3.1	6.3

sera

,725

Table II

Antibacterial Activity Against Anaerobic Gram-Negative Bacteria

		MIC (μg/ml)				
Organism	SC#	SQ 26,726	Cefuroxime	Cefmetazole	Cefotaxime	Gentamicin
B. fragilis	9005	>100.0	>100.0	25	>100.0	>100.0
B. fragilis	9844	100.0	>100.0	6.3	25.0	>100.0
B. fragilis	10,277	>100.0	>100.0	12.5	>100.0	>100.0
B. fragilis	10,278	>100.0	>100.0	50.0	100.0	>100.0
B. fragilis	10,279	>100.0	>100.0	6.3	>100.0	>100.0
B. fragilis	10,281	>100.0	>100.0	6.3	50.0	>100.0
B. fragilis	11,085	>100.0	>100.0	6.3	>100.0	>100.0
B. fragilis	11,086	>100.0	100.0	6.3	100.0	>100.0
H. vaginalis	8568	6.3	0.1	<0.05	<0.1	6.3
H. vaginalis	9640	6.3	0.1	<0.05	<0.1	6.3
F. necrophorum	10,338	>100.0	0.2	<0.05	0.2	12.5

Inoculum = 5×10^5 CFU; Medium = DST Agar (Oxoid) + 5% Sheep Blood

Table 12 Antibacterial Activity Against Recent Clinical Isolates of Gram-Negative Pcds

Organism	SC #	SQ 26,726	Ceftazidime	Cefotaxime	Cefuroxime	Gentamicin
Shigella	12376	<0.05	<0.05	<0.05	1.6	1.6
	12377	<0.05	0.1	<0.05	1.6	3.1
Acinetobacter	12243	6.3	-	3.1	-	1.6
	12244	50	-	12.5	-	0.8
	12409	50	6.3	25	100	1.6
	12410	100	6.3	25	>100	1.6
	12538	50	12.5	25	50	0.4
Salmonella	12370	0.1	0.4	0.1	6.3	6.3
Morganella	12511	<0.05	<0.05	<0.05	12.5	0.4
Achromobacter	12225	>100	-	100	-	50
Bordetella bronchiseptica	2798	>100	25	100	-	6.3
	9320	>100	25	100	-	12.5
Brucinella canimelitis	10954	1.6	0.1	0.2	-	0.8
	11055	1.6	0.1	0.2	-	0.8
	5348	0.4	<0.05	<0.05	-	1.6
Ephia alvei	3194	12.5	12.5	25	-	3.1
	10177	12.5	6.3	3.1	-	1.6
Moraxella bovis	8125	0.2	0.1	<0.05	-	0.8
Ps. maltophilia	11573	3.1	-	1.6	100	3.1
Alcaligenes faecalis	3407	50	-	0.8	50	1.6
	10850	50	-	1.6	50	1.6
Yersinia enterocolitica	10765	0.2	-	<0.05	0.8	0.8
Edwardsiella tarda	9246	<0.05	-	<0.05	0.2	0.8
Vibrio parahemolyticus	9853	3.1	-	0.1	6.3	1.6

DRUG CONTROL REVIEW NOTES

1. TYPE ☒ 5 ☐ IND ☐ 6

2. NO. 50-580

3. SPONSOR
E.R. Squibb & Sons Inc.
P.O. Box 191

4. ADDRESS
New Brunswick, N.J.
ATTN: Norman Lavy, M.D., Vice President
~~Drug Regulatory Affairs~~

5. SUBMISSIONS REVIEWED

a. ORIGINAL DATED

b. AMENDMENTS DATED
See attached

5a. PROVIDING FOR
Manufacturing controls

6. NAME(S)
a. TRADE
AZACTAM

b. NON-PROPRIETARY
Aztreonam for Injection

c. CHEMICAL

d. ESTAB

e. USAN

f. WHO

7. STRUCTURAL FORMULA

8. DOSAGE FORM

9. ☐ RX ☐ OTC

10. FAMILY OR TYPE OF DRUG

11. RELATED NDA, IND, MF, FORM 5's

12. REMARKS
Amendment dated February 15, 1984 responds to all deficiencies existing in the Form 5 application.

13. CONCLUSIONS
Controls are adequate.

14. DATE REVIEWED 6/7/84

15. REVIEWER Joan M. Eckert

FORM FDH-1742 5/65

COPY TO: 1. Original IND HFN-815, HFN-815/CSO, HFN-178
2. Duplicate IND HFN-235
3. Triplicate IND

RD: init. by RNorton/6/15/84

HFN-815/JMEckert/7/19/84/dv

1/5/83 Corrections for sterility test

1/16/84 Animal studies

1/17/84 New synthesis - Process C

2/1/84 Response to telephone conversation of 1/23/84 concerning solubility

2/15/84 Manufacturing controls

4/19/84 Animal studies

4/23/84 Sterility test corrected 1/5/83 should not have been changed.

DRUG CONTROL REVIEW NOTES

1. TYPE ☐ IND ☒ 6

2. NO. 50-580

3. SPONSOR
E.R. Squibb & Sons, Inc.
Box 191

4. ADDRESS
New Brunswick, N.J. 08903
ATTN: Norman Lavy, M.D.

5c. PROVIDING FOR
Microbiology
Corrections for clinical studies

5. SUBMISSIONS REVIEWED

a. ORIGINAL DATED

b. AMENDMENTS DATED
7/12/84

7/16/84

6. NAME(S)

a. TRADE
Azactam

b. NON-PROPRIETARY
Aztreonam

c. CHEMICAL

d. ESTAB

e. USAN

f. WHO

7. STRUCTURAL FORMULA

8. DOSAGE FORM
Injectable: 0.5, 1.0 + 2.0 gram container

9. ☒ RX ☐ OTC

10. FAMILY OR TYPE OF DRUG
Antibiotic

11. RELATED NDA, IND, MF, FORM 8'S

12. REMARKS

See attached Remarks.

13. CONCLUSIONS

Controls remain adequate.

14. DATE REVIEWED
8/2/84

15. REVIEWER
Joan M. Eckert

FORM FDH-1742
5/65

COPY TO: 1. Original IND HFN-815, HFN-815/CSO, HFN-178
2. Duplicate IND
3. Triplicate IND HFN-235
HFN-815/JMEckert/8/6/84/dv

R/D: init. by RNorton/9/4/84

9/4

12. Remarks:

The July 16, 1984 amendment includes a report from the Clinical Microbiology Institute Inc. This was an evaluation of control parameters for the control organisms used in the susceptibility testing of aztreonam. Only Escherichia coli and Pseudomonas aeruginosa will be used as control organisms. Staphylococcus aureus will not be used as a control organism because of the lack of aztreonam activity against this organism.

Nine laboratories each tested samples from three lots of 30/ug aztreonam discs using a different lot of Mueller/Hinton agar, 50 tests with each organism. (150 zone measurements) In addition, each laboratory performed 5 separate tests with a cross-over lot of Mueller Hinton agar (15 zone measurements).

The quality control parameters recommended are as follows:

Zone diameters (mm)	E. coli (ATCC 25922)	. aeruginosa (ATCC 27853)
Observed range[1]	27 - 38	22 - 32
Mean	32.1	26.2
Mean ± 2SD	28.4 - 35.8	22.8 - 29.6
Median	32	26
Proposed limits[2]	28 - 36	23 - 29

[1] Based on 1350 determinations (150 from each of 9 laboratories)

[2] Median ± half of the median of the ranges for the nine laboratories.

MICROBIO

REVIEW

50-580

MICROBIOLOGY REVIEW OF LABELING

Date submitted: 12/28/83

The labeling was not filed with the original submission which contained only the manufacturing controls and preclinical data. Therefore, the original manufacturing controls review did not include a review of the labeling. Vial and package labels are adequate.

PACKAGE INSERT

The Microbiology section of the package insert is satisfactory.

Data from eleven clinical studies were filed.
Study 18,554-10 compared spectinomycin with aztreonam in acute uncomplicated gonorrhea infections.

There were seven microbiological failures. Of these seven, three were also clinical failures. The MIC's for the seven microbiological failures were: 0.25u/ml*; 0.25u/ml; 0.25u/ml; 0.03u/ml*; 0.015u/ml* and 0.03u/ml. One isolate was not tested.

* Microbiological and clinical failures.

There were eight clinical failures in which the organism was eradicated. The MIC's for these isolates were as follows: 0.125u/ml; 0.125u/ml; 0.06u/ml; 0.03u/ml; 0.007u/ml; 0.06u/ml; 0.25u/ml; and 0.03u/ml.

The total number of isolates from all sites was 264. Sixty one percent (162) were =0.1, 23%(60) were between 0.2 and 0.4, 2.2%(6) were between 0.5 and 1.0, 0.7%(2) were between 2 and 4. Thirty four not reported.

The susceptibility criteria for MIC testing in the labeling is:

≤ 8 Susceptible
16 Intermediate
≥ 32 Resistant

The proposed criteria are not applicable to Neisseria gonorrhea. As shown in the above data, the isolates involved in the microbiological and clinical failures do not fall into the resistant category. The MIC of the organism does not appear to be predictive of the outcome of the disease.

Six of the eleven studies were treatment of urinary tract infections: 18,554-13; 18,554-14; 18,554-15; 18,554-27; 18,554-28 and 18,554-31. Study 18,554-31 included infection due to multi-drug resistance.

In each clinical failure, at least one of the organisms was a microbiological failure as well.

The following table includes microbiological clinical failures and the organisms involved:

In all of the following studies, an aerobic, gram-negative organism susceptible to aztreonam had to be isolated pretherapy for the patient to be evaluable.

Protocol #	Micro failures	Clin. failures	Part. failures	E. coli	P. aerug.	K. pneum.	P. mirab.	S. marces.	E. aerog.	Ent. cloacae	P. stuartii
13	9	2*	1	7	2						
14	18	1*	1	8	3	2	2	1	1	1	
15	9	4*	4	9							
27	5	0		2	2	1					
28	8	0		3	3				1		1
31	8	3**		1	6	1					
	57	10		30	16	4	2	1	2	1	1

* E. coli

** P. aeruginosa

There was a total of 393 evaluable patients treated with aztreonam, 57 microbiological failures (14.5%). Of the 57 microbiological failures, 10 were clinical failures and 20 were partial failures. Fifty three percent of the microbiological failures were either clinical or partial failures.

Six of the ten clinical failures were due to single gram negative pathogens.

One was single gram negative pathogen with a superinfection due to S. faecalis. In 3 others, S. faecalis was present with gram neagative organisms.

In study, 18,554-11, tobramycin was compared with aztreonam in the treatment of lower respiratory tract infections due to gram negative organisms.

Of 98 evaluable patients, there was six clinical failures. In 2 of the failures, single pathogens were isolated. One clinical failure was due to Klebsiella oxytoca, 30 mm zone, and an MIC of 3. In the second failure Pseudomonas aeruginosa was isolated, zone of 29 mm. On later testing, a P. aeruginosa isolated from the same patient tested as resistant.

The four other cases were mixed infections with gram positive organisms present. One of these failures was due to super infection of Pseudomonas fluorescens, zone size 13 mm. The P. fluorescens was not isolated until Day 7 of treatment. A second isolate, P. aeruginosa, from the same patient was susceptible pre Rx and may have become resistant (Vari 287). Details follow.

There were fourteen microbiological failures:

9 P. aeruginosa - 2 clinical failures
4 partial cures
3 cures

1 E. coli Clinical cure
1 K. oxytoca Partial cure
2 E. cloacae Clinical cures
1 Veillonella Partial cure

Of the fourteen microbiological failures, six remained susceptible throughout the study. The remaning eight failures may have been due to the development of resistance. However, this has not been definitely established. The firm recognizes resistance in four of the pathogens*. Data for each of the eight failures is as follows:

Brusch 061 Microbiological failure Partial cure Pneumonia
6224

20 days of treatment 8/10/82-8/30/82
60 grams of drug administered

P. aeruginosa			P. maltophilia		
8/10/82	28 mm	Sputum	8/14/82	28 mm	Sputum
8/11/82	24 mm	"	8/18/82	30 mm	No source given
8/14/82	S	"	8/26/82	S	" " "
8/18/82	S	"	9/15/32	R	" " "
8/23/82	S	"			
8/26/82	S	"			

Firm considers the P. aeruginosa a failure because it was not eradicated. The P. maltophilia was the cause of a superinfection.

Ga ner 052* Microbiological failure Clinical cure Pneumonia
6227

11 days of treatment 2/24/83-3/5/83
60 grams of drug administered
Multiple pathogens, P. aeruginosa considered a failure.

P. aeruginosa				E.R. Squibb results	
2/24/83	22 mm	6.25 µg/ml	Sputum	Pre Rx 22 mm	6.23 µg/ml
2/26/83	26 mm	6.25 µg/ml	"	Post Rx 6 mm	>50
3/1/83	12 mm	>50 µg/ml	"		
3/3/83	13 mm	50 µg/ml	"		
3/6/83	6 mm	>50 µg/ml	"		

Ramirez-Ronda 057* Microbiological failure Clinical failure
6228

12 days of treatment 2/4/83-2/16/83
72 grams of drug administered

P. aeruginosa				E.R. Squibb results	
2/2/83	29 mm		sputum	Pre Rx	29 mm -
2/6/83	No good		"	Day 8	0 mm 128 ug/ml
2/11/83	0 mm	> 128 ug/ml	"		

Vari 213 Microbiological failure Clinical cure
7614 Multiple pathogens - Enterobacter cloacae is considered a failure but is not considered a pathogen.

9 days of treatment 8/18/82-8/26/82
46 grams of drug administered

E. cloacae			
8/16/82	S	Considered a pathogen	Sputum
8/25/82	R	Not considered a pathogen	Sputum

216* Microbiological failure Partial cure Pneumonia

13 days of treatment 10/1/82-10/13/82
39 grams of drug administered

P. aeruginosa and K. oxytoca were isolated

P. aeruginosa

10/1/82	25 mm	Sputum
10/7/82	13 mm	Sputum
10/13/82	R	"
10/17/82	R	"

E.R. Squibb results

Pre Rx	25 mm	25 µg/ml
Post Rx	R	25 µg/ml

K. oxytoca was susceptible

287 Microbiological failure Clinical failure Pneumonia
Multiple infection - P. aeruginosa, P. fluorescens, K. pneumoniae and S. aureus. P. aeruginosa is considered a failure.

8 days of treatment 11/30/82-12/7/82
30 grams of drug administered

P. aeruginosa	11/30/82	20 mm	Trach.
	12/6/82	23 mm	"
	12/26/82	23 mm	"
	1/3/83	R	"

301* P. fluorescens 12/6/82 13 mm Trach. Superinfection
Microbiological failure Clinical cure Pneumonia
Mixed infection - P. aeruginosa and Acinetobacter calcoaeticus, P. aeruginosa is considered the failure.

9 days of treatment 4/9/83-4/17/83
36 grams of drug administered

P. aeruginosa

4/7/83-	24mm	12.5 µg/ml
4/14/83	10mm	>25 µg/ml
4/18/83	12mm	>25 µg/ml

E.R. Squibb Results

Pre Rx	24 mm	12.5 µg/ml
Post Rx	12mm	>25 µg/ml

Weinstein 437 Microbiological failure Clinical cure Empyema
6401 Multiple pathogens - Enterobacter cloacae is considered the failure

23 days of treatment 1/23/83-2/14/83
66 grams of drug administered

E. cloacae

1/23/83	≤.5	µg/ml	Trach
1/25/83	≤.5	µg/ml	Trach & Pleural fluid
1/27/83	16	µg/ml	Trach
1/29/83	32	µg/ml	"
2/1/83	32	µg/ml	"
2/5/83	32	µg/ml	Bronch
2/8/83	16	µg/ml	Trach
2/12/83	32	µg/ml	Trach
2/15/83	32	µg/ml	Sputum

P. fluorescens Treatment emergent

2/5/83 32 µg/ml Not repeated Not considered a pathogen

In Study 18,554-16, aztreonam was evaluated in the treatment of serious infections due to gram-negative organisms. Urinary tract, skin and skin structures, lower respiratory tract, septicemia, intrabdominal, bones and/or joints and obstetric and gynecologic infections were included in this study.

According to the patient summary sheets, there were 97 microbiological failures as follows:

55 P. aeruginosa
- 44 Susceptible
- 4 Intermediate
- 1 Resistant
- 6 Unable to determine from investigators report

Of the 55 microbiological failures, 12 were isolated from patients who were clinical failures and 24 from partial cures.

16 E. coli

One isolate may have become resistant. There were 3 clinical failures and 4 partial cures involving these organisms.

4 K. pneumoniae
- 2 clinical failures

4 P. mirabilis
- 2 partial cures

3 Enterobacter cloacae

1 clinical failure and 1 partial cure

Sabath 4232 011 Microbiological failure Partial cure Abscess

15 days of treatment 9/30/82-10/14/82
Total dose 96 grams

P. aeruginosa — E.R. Squibb results

9/27/82 20.3mm 16 µg/ml abdominal fluid Pre Rx 24mm 25 µg/ml
9/27/82 25mm 16 µg/ml abdominal drainage Post Rx 20.3mm 12.5 µg/ml
10/4/82 24.9mm 32 µg/ml abdominal drainage
10/12/82 13.5mm 64 µg/ml abdominal wound
10/15/82 12.3mm 64 µg/ml Drainage
10/15/82 13mm 32 µg/ml Swab, abdomen

Nolen 6449 013 Microbiological failure Partial cure UTI & Pneumonia

12 days of treatment 2/3/83-2/14/83
Total dose of drug administered 38 grams

	Enterobacter aerogenes				K. pneumoniae
2/3/83	14 mm	.5 Urine	12 mm	16 µg/ml Sputum	24 mm ∠.5 Sputum
2/6/83	No growth				No growth
2/9/83	No growth				No zone >64
2/12/83	No growth				No growth
3/1/83					Urine 23 mm

Schalkhauser 6453 013 Microbiological failure Partial cure UTI

7 days of treatment 4/5/83-4/11/83
Total dose administered - 21 grams

E. coli

4/5/83 24 mm
4/15/83 16 mm
5/9/83 12 mm

Sereni 7551 007 Microbiological failure Partial cure Pneumonia

17 days of treatment 5/3/83-5/19/83
Total dose administered 25 grams

2 Enterobacter aerogenes

1 resistant which resulted in a partial cure
1 susceptible which became resistant and resulted in a clinical failure

2 S. marcescens

1 clinical cure
1 partial

1 S. liquifaciens

Clinical failure

1 P. fluorescens

Partial cure

There was a total of 26 clinical failures. Two clinical failures each involved one of the 2 resistant isolates, E. aerogenes (6449-013) and P. aeruginosa (7533-001). In 16 of these clinical failures, at least one pathogen was not eradicated. In eight of these clinical failures, all pathogens were eradicated.

Nine of the microbiological failures may have developed resistance, however this has not been definitely established. Details for six are as follows:

Sabath 010 Microbiological failure Clinical failure Cellulitis
4232 Osteomyelitis

18 days of treatment 9/27/82-10/14/82
Total dose of drug administered 134 grams
Cefamandole 9/21/82-9/27/82
Gentamicin 9/25/82-9/27/82

P. aeruginosa			E.R. Squibb results		
9/21/82	24.3 mm.	4 µg/ml	Pre RX	26 mm	6.3 µg/ml
9/29/82	24 mm	8 µg/ml	Post RX	6 mm	>50 µg/ml
10/11/82	6 mm	12.8 µg/ml			
10/14/82	6 mm	64 µg/ml			

The susceptibility to gentamicin changed from susceptible to intermediate. It was tested on the same days

P. maltophilia

4/26/83 S
5/19/83 R

E.R. Squibb results

Pre Rx S
Post Rx R

Ambrosioni 005 Microbiological failure Partial cure Pneumonia
7/24

10 days treatment 5/23/83-6/1/83
Total dose administrative 70 grams

Pseudomonas sp.

5/23/83 27 mm
5/28/83 15 mm
6/2/83 0 mm

Three of the microbiological failures which appeared to develope resistance were P. aeruginosa isolates. The MIC's seemed to change from 1.25 µg/ml to 50 or 100 µg/ml. But investigator's report is difficult to read. These are "Neu 013, 061, and 080.

Study 18,554-38 is a comparison of aztreonam plus clindamycin with tobramycin plus clindamycin in the treatment of intra-abdominal infections. There were four clinical failures. Two were microbiological failures-5766-101 and 6407-207. In 6407-207 the Pseudomonas aeruginosa may have developed resistance.

Microbiological failure - Clinical failure
11 days of treatment 9/3/82 - 9/13/82
Total dose of drug - 66 grams

P. aeruginosa

9/14/82 20 mm
9/6/82 S
9/15/82 R

Study 18,554-41 is a comparison of aztreonam plus clindamycin in the treatment of obstetric and gynecologic infections.
There were no microbiologic or clinical failures.

Conclusions

Excluding the gonorrhea study, there was a total of 971 evaluable patients. There were 46 clinical failures and 171 microbiological failures.

Forty four (4.5%) of the clinical failures were not predicted by firm's susceptibility criteria.

E. coli was the organism isolated more times than any other pathogen, 387 isolates including single and multiple infections. The next largest number of organisms isolated was P. aeruginosa, 245. However, the largest number of microbiological failures, 31, was in the Pseudomonas group. There were 46 microbiological failures in the E. coli group.

Joan M. Eckert, 8/9/84

cc: Orig. Form 5 5-580
HFN-815, HFN-815/CSO
HFN-178, HFN-235
HFN-815/JMEckert/8/10/84/dv

ANALYTICAL METHODS STABILITY CONTROL

Autobac 1

7. PRODUCT DESCRIPTION

III. Disposables

Pfizer

SECTION 1

A full list of the articles used as components of the drug. This list should include all substances used in the fermentation, synthesis, extraction, purification or other method of preparation of any antibiotic and in the preparation of the finished dosage form, regardless of whether they undergo any change or are removed in the process. Each substance should be identified by its established name, if any, or complete chemical name, using structural formulas when necessary for specific identification. If any proprietary preparation is used as a component, the proprietary name should be followed by a complete quantitative statement of composition. Reasonable alternatives for any listed substance may be specified.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

I. Ampicillin Elution Susceptibility Testing Disks

A. Ampicillin trihydrate, manufactured and assayed by Bristol Laboratories, Syracuse, New York, or equivalent.

Filter paper

II. Bacitracin Elution Susceptibility Testing Disks

A. Bacitracin

Filter paper

III. Carbenicillin Elution Susceptibility Testing Disks

A. Carbenicillin

Pfizer

SECTION 1 (continued)

IV. Cephalothin Elution Susceptibility Testing Disks

Cephalothin, sodium, manufactured and assayed by Eli Lilly and Company, Indianapolis, Indiana, or equivalent

Filter paper

V. Chloramphenicol Elution Susceptibility Testing Disks

Chloramphenicol, manufactured and assayed by Parke-Davis and Company, Detroit, Michigan, or equivalent

Filter paper

VI. Clindamycin Elution Susceptibility Testing Disks

Clindamycin, manufactured and assayed by The UpJohn Company, Kalamazoo, Michigan, or equivalent

Filter paper

VII. Colistin Elution Susceptiblity Testing Disks

Colistin sulfate, manufactured and assayed by Warner-Chilcott Company, Morris Plains, New Jersey, or equivalent

Filter paper

VIII. Doxycycline Elution Susceptiblity Testing Disks

Doxycycline

Pfizer

Filter paper

IX. Erythromycin Elution Susceptibility Testing Disks

Erythromycin, manufactured and assayed by Eli Lilly and Company, Indianapolis, Indiana, or equivalent

Filter paper

X. Gentamicin Elution Susceptibility Testing Disks

Gentamicin sulfate, manufactured and assayed by Schering Corporation, Union, New Jersey, or equivalent

Filter paper

XI. Kanamycin Elution Susceptibility Testing Disks

Kanamycin sulfate, manufactured and assayed by Bristol Laboratories, Syracuse, New York, or equivalent

Filter paper

XII. Methicillin Elution Susceptibility Testing Disks

Methicillin, sodium, manufactured and assayed by Bristol Laboratories, Syracuse, New York, or equivalent

Filter paper

Pfizer

XIII. Nalidixic Acid Elution Susceptibility Testing Disks

Nalidixic Acid, manufactured and assayed by Winthrop Laboratories, New York, New York, or equivalent

Filter paper

XIV. Neomycin Elution Susceptibility Testing Disks

Neomycin sulfate

Filter paper

XV. Nitrofurantoin Elution Susceptibility Testing Disks

Nitrofurantoin, manufactured and assayed by H. Reisman Corp., New Hyde Park, New York, or equivalent

Filter paper

XVI. Novobiocin Elution Susceptibility Testing Disks

Novobiocin, sodium, manufactured and assayed by The Upjohn Company, Kalamazoo, Michigan, or equivalent

Filter paper

XVII. Oleandomycin Elution Susceptibility Testing Disks

Oleandomycin phosphate

Filter paper

XVIII. Penicillin G Elution Susceptibility Testing Disks

Penicillin G

Filter paper

XIX. Polymyxin B Elution Susceptibility Testing Disks

Polymyxin B sulfate, manufactured and assayed by Burroughs-Wellcome Company, Tuckahoe, New York, or equivalent

Filter paper

XX. Streptomycin Elution Susceptibility Testing Disks

Streptomycin sulfate

er paper

XXI. Tetracycline Elution Susceptibility Testing Disks

Tetracycline hydrochloride

Filter paper

XXII. Vancomycin Elution Susceptibility Testing Disks

Vancomycin hydrochloride, manufactured and assayed by Eli Lilly and Company, Indianapolis, Indiana, or equivalent

Filter paper

Pfizer

SECTION I (continued)

XXIII. Lincomycin Elution Susceptibility Testing Disks

Lincomycin, HCl. manufactured and assayed by the Upjohn Company, Kalamazoo, Michigan or equivalent.

Filter paper

Pfizer

SECTION I (continued)

Except where otherwise noted, all bulk antibiotics utilized as components of these drugs are manufactured and assayed by Pfizer Inc., New York, New York.

It is recognized that disks utilizing Nalidixic Acid and Nitrofurantoin are not subject to certification. The data included here for these products are meant for information only.

Pfizer Diagnostics currently manufactures similar certified susceptibility disks meant for use in the standardized Kirby-Bauer agar diffusion test under the following Antibiotic Form 6 numbers and dates:

Antibiotic	Disk Potency	FDA Form 6 Number	Date Approved
Ampicillin	2 and 10 mcg	60-974	June 1964
Bacitracin	2 and 10 U	60-975	March 1961
Carbenicillin	50 mcg	61-447	March 1971
Cephalothin	30 mcg	60-977	February 1965
Chloramphenicol	5 and 30 mcg	60-978	March 1961
Clindamycin	2 mcg	61-333	December 1970
Colistin	2 and 10 mcg	60-981	June 1962
Doxycycline	5 and 30 mcg	60-984	January 1968
Erythromycin	2 and 15 mcg	60-985	March 1961
Gentamicin	10 mcg	60-912	September 1969
Kanamycin	5 and 30 mcg	60-986	April 1965
Methicillin	5 mcg	60-989	March 1962
Neomycin	5 and 30 mcg	60-991	March 1961
Novobiocin	5 and 30 mcg	60-992	March 1961
Oleandomycin	2 and 15 mcg	60-994	March 1961
Penicillin	2 and 10 U	60-997	March 1961
Polymyxin B	50 and 300 U	60-999	March 1961
Streptomycin	2 and 10 mcg	61-000	March 1961
Tetracycline	5 and 30 mcg	61-002	March 1961
Vancomycin	5 and 30 mcg	61-003	April 1965
Lincomycin	2 mcg	60-987	April 1965

Pfizer

SECTION 2

A full statement of the composition of the drug. The statement shall set forth the name and amount of each ingredient, whether active or not, contained in stated quantity of the drug in the form in which it is to be distributed, as for example, amount per tablet or per milliliter, and a batch formula representative of that to be employed for the manufacture of the finished dosage form. All components should be included in the batch formula regardless of whether they appear in the finished product. Any calculated excess of an ingredient over the label declaration should be designated as such and per cent excess shown. Reasonable variations may be specified.

Elution Susceptibility Testing Disks consist of filter paper disks impregnated with appropriate quantities of an antibiotic drug dissolved in an appropriate solvent with or without buffer. The amounts of antibiotic, solvent and buffer, if required, are calculated for each batch of elution disks depending upon drug activity and batch size involved. For details as to the manner of performing these calculations, please refer to the Master Production Sheet-Sensitivity Disks, exhibited in conjunction with Section 3 f of this application.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

I. Ampicillin Elution Susceptibility Testing Disks

A. Low concentration disk

Ampicillin trihydrate, 0.22 mcg/disk

B. High concentration disk

Ampicillin trihydrate, 3.6 mcg/disk

II. Bacitracin Elution Susceptibility Testing Disks

Bacitracin, 18 U/disk

Pfizer

SECTION 2 (continued)

III. Carbenicillin Elution Susceptibility Testing Disks

Carbenicillin, 120 mcg/disk

IV. Cephalothin Elution Susceptibility Testing Disks

Cephalothin, 15 mcg/disk

V. Chloramphenicol Elution Susceptibility Testing Disks

Chloramphenicol, 4 mcg/disk

VI. Clindamycin Elution Susceptibility Testing Disks

Clindamycin, 2 mcg/disk

VII. Colistin Elution Susceptibility Testing Disks

Colistin sulfate, 13 mcg/disk

VIII. Doxycycline Elution Susceptibility Testing Disks

A. Low concentration disk

Doxycycline, 0.5 mcg/disk

B. High concentration disk

Doxycycline, 1.6 mcg/disk

Pfizer

SECTION 7 (continued)

IX. Erythromycin Elution Susceptibility Testing Disks

Erythromycin, 2.5 mcg/disk

X. Gentamicin Elution Susceptibility Testing Disks

Gentamicin sulfate, 9 mcg/disk

XI. Kanamycin Elution Susceptibility Testing Disks

Kanamycin sulfate, 22 mcg/disk

XII. Methicillin Elution Susceptibility Testing Disks

Methicillin, sodium, 5 mcg/disk

XIII. Nalidixic Acid Elution Susceptibility Testing Disks

Nalidixic Acid, 15 mcg/disk

XIV. Neomycin Elution Susceptibility Testing Disks

Neomycin sulfate, 24 mcg/disk

XV. Nitrofurantoin Elution Susceptibility Testing Disks

Nitrofurantoin, 15 mcg/disk

XVI. Novobiocin Elution Susceptibility Testing Disks

Novobiocin, sodium, 2.5 mcg/disk

N 50580 -6

Pfizer

SECTION 2 (continued)

XVII. Oleandomycin Elution Susceptibility Testing Disks

Oleandomycin phosphate, 6 mcg/disk

XVIII. Penicillin G Elution Susceptibility Testing Disks

Penicillin G, 0.2 U/disk

XIX. Polymyxin B Elution Susceptibility Testing Disks

Polymyxin B sulfate, 12.5 U/disk

XX. Streptomycin Elution Susceptibility Testing Disks

Streptomycin sulfate, 20 mcg/disk

XXI. Tetracycline Elution Susceptibility Testing Disks

A. Low concentration disk

Tetracycline hydrochloride, 0.5 mcg/disk

B. High concentration disk

Tetracycline hydrochloride, 1.2 mcg/disk

XXII. Vancomycin Elution Susceptibility Testing Disks

Vancomycin hydrochloride, 10 mcg/disk

XXIII. Lincomycin Elution Susceptibility Testing Disks

Lincomycin, 2.4 mcg / disk

Pfizer

SECTION 3

A complete description of the methods and processes used in manufacturing, packing and labelling of the drug to preserve its identity, strength, quality and purity in conformity with good manufacturing practices including:

(Pfizer)

SECTION 3a

Name and location of each plant conducting the operations.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

All Antibiotic Elution Susceptibility Testing Disks are manufactured, assayed for potency, labelled and controlled at our plant at 199 Maywood Avenue, Maywood, New Jersey 07607. This plant is registered with the FDA as a Drug Establishment under Number 22-1959G on FD Form 1597. These facilities were most recently inspected by FDA in February 1973.

Pfizer

SECTION 3b

Whether or not each lot of raw materials is given a serial number to identify it, and the use made of such numbers in subsequent plant operations.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

All antibiotic elution susceptibility testing disks containing certifiable antibiotics are manufactured and certified in compliance with the regulations set forth for similar disks designed for use in agar diffusion tests in 21 CFR 147.1 and 147.2. Equivalent manufacturing procedures and controls are applied to the production of elution susceptibility testing disks containing non-certifiable antimicrobial agents.

Strips of filter paper are pre-printed with the appropriate antibiotic code symbol and the designation "ep", "en", or "ea" in order to identify the contained antibiotic/antimicrobial and to distinguish the disk from those used for standard susceptibility testing carried out by agar diffusion methods such as the Kirby-Bauer procedure.

The paper used is

. pre-tested to conform to the regulations set forth in 21 CFR 147.1 (d). On release by our Quality Control Laboratory, each lot of bulk paper, identified by the manufacturer's assigned batch number, is released for printing and cutting.

All antibiotics used for impregnation are obtained from reputable manufacturers who have on file with us appropriate Antibiotic Forms 4. Their assay values are accepted if confirmed by our own assay procedure.

Released materials are identified by the manufacturer's assigned batch lot number. This identifying number appears on each Batch Production Control Sheet for all disk lots made from it. Accountability for usage and disposition of all antibiotics and antimicrobials is maintained via Bulk Drug Inventory Control Sheets.

Solvents and buffers used are ACS reagent grade materials on which appropriate identity tests are carried out by our Quality Control Laboratory.

Identifying manufacturer's lot numbers appear on Batch Production Control Sheets for all disk lots employing the subject materials.

Distilled water is used when required. Its purity is monitored by our Quality Control Laboratory.

Pfizer

SECTION 3e

Precautions to assure proper identity, strength, quality and purity of the raw materials, whether active or not, including the specifications for acceptance and methods of testing for each lot of raw material used in the fermentation, synthesis, extraction, and purification of the drug and for each ingredient used in the manufacture of the drug that is to be dispensed.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

Components

Standards of acceptance are established for all components (active and inactive). These specifications are designed to ascertain that components of the disks are of adequate quality, possess the necessary attributes of identity, quality and purity and conform to the requirements of established compendia such as the U. S. P. or N. F., when applicable.

Prior to use, the components used in manufacture are evaluated and released by the Quality Control Laboratory.

Pfizer

SECTION 3d

If it is a drug produced by fermentation.

Not applicable.

Pfizer

SECTION 3e

If it is a drug that is synthesized by chemical processes, a detailed description of each chemical reaction with graphic formulas used to produce the drug, including the names and amounts of all substances used in the process.

Not applicable.

Pfizer

SECTION 3C

Method of preparation of the master formula records and individual batch records and manner in which these records are used.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

Control Procedures

Control procedures are described in the enclosed copy of Master Production Sheet-Sensitivity Disks (Revision number 2, January 13, 1973).

A batch lot number is assigned to each batch of impregnated strips. This number is obtained from a master bound book which lists all disks made in chronological order. The numbers in this master book are listed in ascending numerical sequence. The production sheets for a specific batch are marked with the designated batch lot number and the book is marked with the drug name, potency and date of manufacture. This batch lot number is likewise marked on the racks containing the impregnated strips which are being dried, the containers in which dried impregnated strips are stored, the Quality Control assay sheets bearing the assay data for the batch, and the boxes in which packages of disks from this batch are stored in quarantine.

After the batch has been certified, and released in writing by Quality Control, the assigned expiration date is entered into the master book. When packages of disks are labelled, the same lot number and expiration date are imprinted onto each label used and appropriate records maintained in the Label Accountability Book.

Each batch lot is fully described and identified on an individual Production Sheet-Sensitivity Disks and on Disk Disposition Sheets (see enclosures in Section 3q).

Revision Number 2, January 13, 1973

July 18, 1970

MASTER PRODUCTION SHEET-SENSITIVITY DISKS

Instructions for Preparation of Batch Production Sheets and Disks

Do Not Alter or Modify This Form Without Written Authorization by Technical Manager

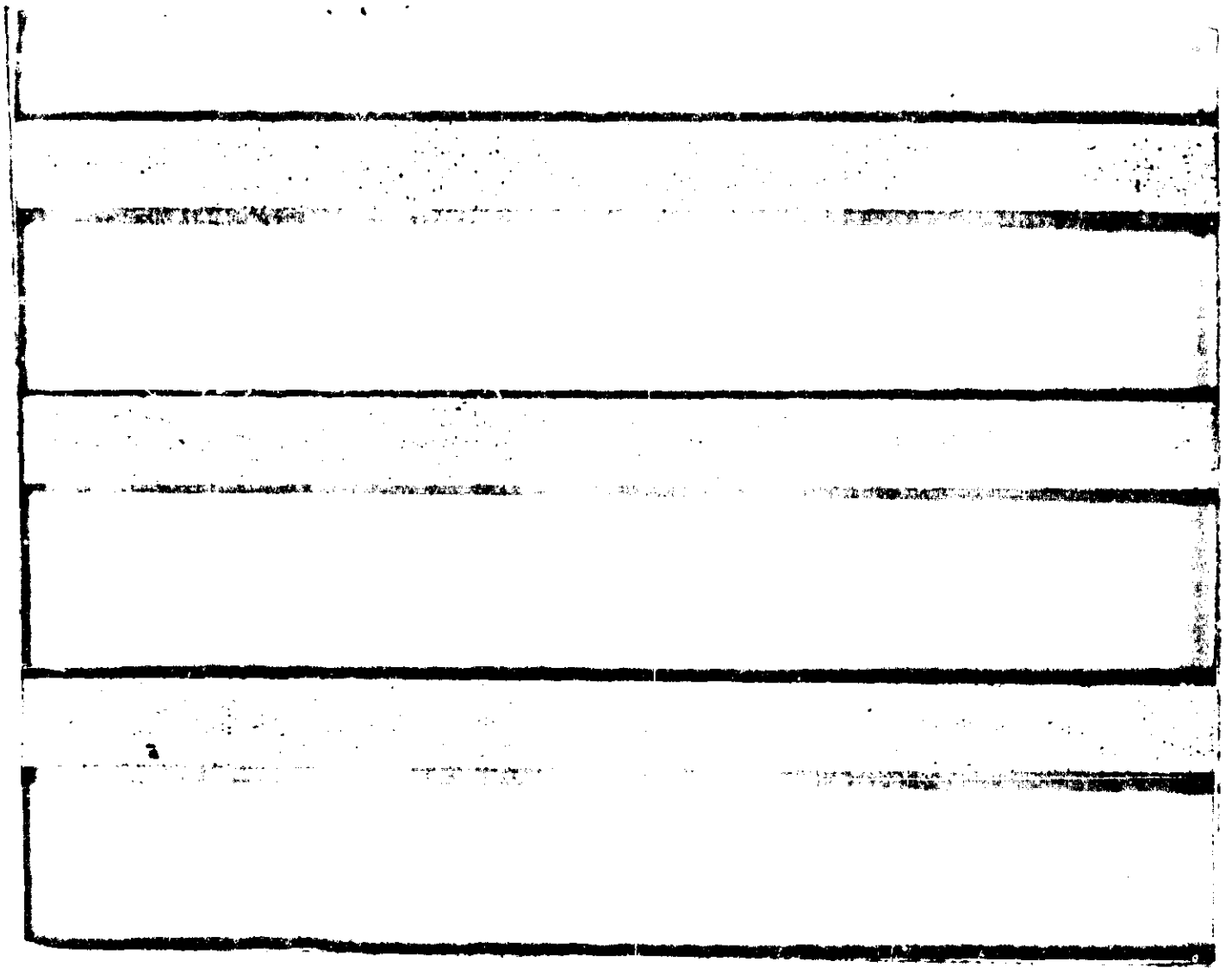
Revision Number 2, January 13, 1973

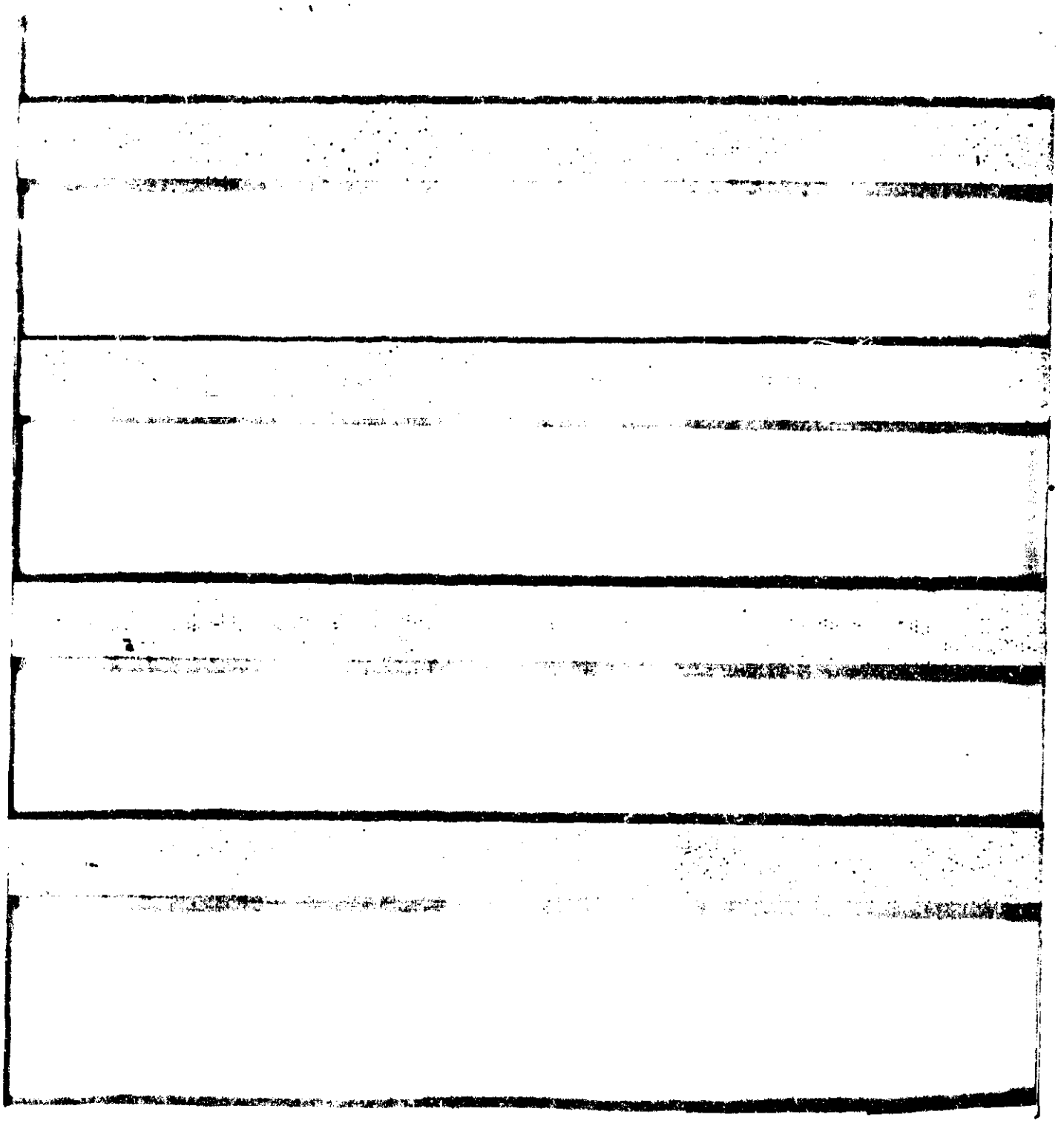
July 18, 1970

MASTER PRODUCTION SHEET-SENSITIVITY DISKS

Instructions for Preparation of Batch Production Sheets and Disks

Do Not Alter or Modify This Form Without Written Authorization by Technical Manager





Pfizer

SECTION 3g

Number of individuals checking weight or volume of each individual ingredient entering into each batch of the drug.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

Number of Individuals Checking Ingredients

As indicated under Section 3f, two individuals check the weight or volume of each ingredient entering into each batch or the drug.

Section 3h

Whether or not the total weight or volume of each batch is determined at any stage of the manufacturing process subsequent to making up the batch according to the formula card, and at what stage and by whom this is done.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

As indicated under Section 3f, calculations of solution, volumes and expected yields are verified and certified by a second, responsible individual. The number of strips actually made is recorded and is available for comparison with the number initially scheduled.

Pfizer

SECTION 31

At what point in the process the drug is mixed homogeneously and a description of the equipment used for this purpose and its total capacity in terms of pounds, kilograms, gallons or liters of the drug and the maximum quantity of the drug that is mixed in such equipment.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

As described in Section 3f, ingredients are weighed out separately and added to appropriate volumes of solvent(s). Following complete solution of the ingredients, the mixture is mixed to a state of homogeneity with a variable speed electrically driven mixer. A final pH value is taken and recorded on the Batch Control Sheet.

At the present time the maximum batch lot volume handled is approximately [redacted] and is expected to yield a maximum number of strips of approximately

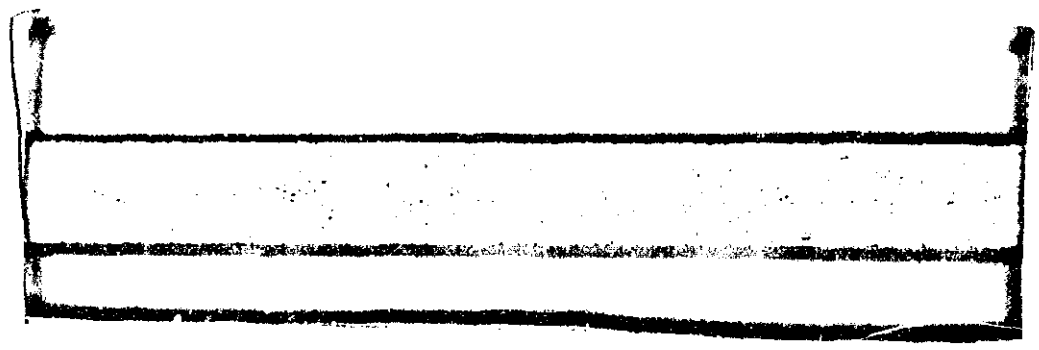
Pfizer

SECTION 3j

A description, where applicable, of all equipment used in the fermentation, synthesis, extraction, purification, filtration, sterilizing, grinding, blending, mixing, tableting, encapsulating, filling, packaging and labelling of the drug.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

In addition to the equipment noted in Sections 3f and 3i, the following equipment may normally be employed:



(Pfizer)

SECTION 3k

If it is a sterile drug, a description of the methods used to insure the sterility of each batch and the controls used for maintaining its sterility, including a detailed description of the sterile areas where the drug is produced and packaged.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

Not applicable.

Pfizer

SECTION 31

Additional procedures employed which are designed to exclude contaminants (e.g., other drug substances, extraneous materials, etc.) and otherwise assure proper control of the product.

[redacted]

SECTION 3.1 - (continued)

[redacted]

Pfizer

SECTION 3m

Adequate information with respect to the characteristics of and the test methods employed for the container, closure, or other component parts of the drug container to insure their suitability for the intended use.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

[redacted]

SECTION 3n

Controls used in the packaging and labelling of each batch to insure the standards of identity, strength, quality and purity of the drug.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

See under Sections 3f and 3l.

Pfizer

SECTION 3o

Precautions to check the total number of finished packages produced from a batch of the drug with the theoretical yield.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

See under Section 3f.

Pfizer

SECTION 3p

Precautions to insure that each lot of the drug is packaged with the proper label and labelling, including provisions for labelling, storage, and inventory control.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

Precautions to Insure that Each Lot of Drug Is Packaged with the Proper Labelling

Label copy is submitted to the following groups for approval:

Sales
Medical
Legal
Quality Control
Drug Regulatory Affairs
Production

Approved labelling is covered by specifications which clearly establish the exact label (or labelling) for the product. Labelling is ordered from the printer under the assigned product number and disk code number. Upon receipt from the printer, labelling is inspected and checked against approved labelling specifications and the labels are placed in Label Stock.

FDA-approved labels specific for each elution disk product are described in Section 6. These labels are distinctively different from those applied to susceptibility disks meant for use in agar diffusion assays such as the Kirby-Bauer method.

A distinctive, FDA-approved package insert is provided for all disks manufactured for use with the Pfizer Diagnostics Autobac 1 Antibiotic Susceptibility Testing System.

(Pfizer)

SECTION 3G

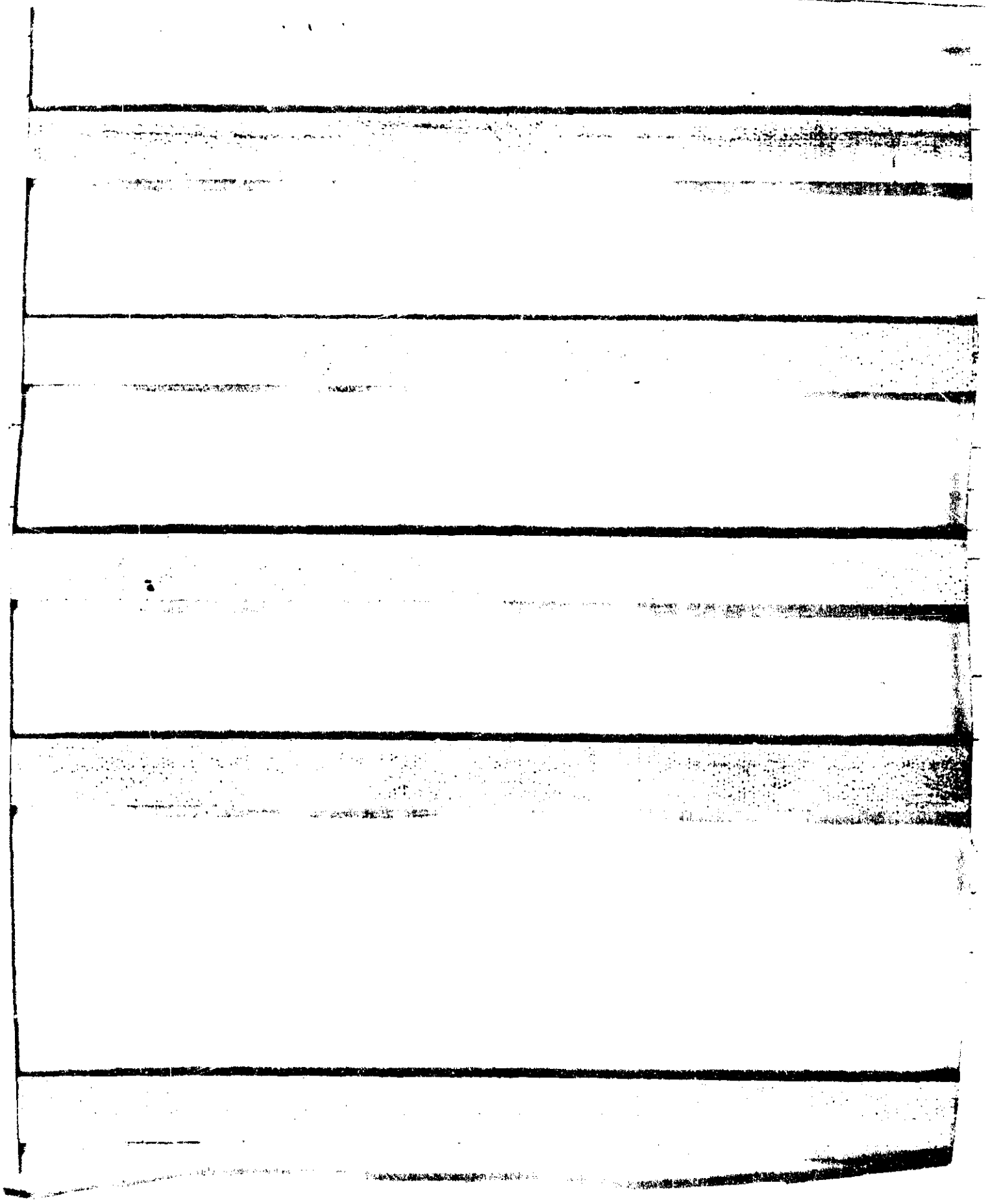
Copies of all printed forms used by the applicant in the manufacture, packaging and labelling of a batch.

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

See attached forms.

PRODUCTION SHEET
SENSITIVITY DISKS

LOT NUMBER



SENSITIVITY DISKS -- WORK IN PROGRESS SHEET

LOT NO.	ITEM	DATE SCHED.	DATE IMPREG.	DATE PACKAGED	DATE SENT TO Q.C.	DATE FDA ASSAY COMPLETE	DATE SENT TO FDA	DATE CERT. RCVD.	DATE CANADA ASSAY	DATE SENT TO CANADA	DATE CANADA APPROVAL	

DRUG INVENTORY SHEET

(NAME)

(Use Separate Sheet for Each Batch of Material)

Date Received_____ Source_____

Lot No._____ Quantity Received_____

Potency_____ Exp. Date_____ Last Assay Date_____

Remarks:

Date	Lot No. of Batch	Amount Used	Balance On Hand	Date	Lot No. of Batch	Amount Used	Balance On Hand

SENSITIVITY DISK - DISPOSITION SHEET LOT NUMBER _____

Name and Potency _____ Symbol _____

Date Sent _____ Date Returned _____

Number Strips Sent _____ Strips Used _____ Strips Destroyed _____

Number Disks Made _____ Disks Used _____ Disks Destroyed _____

Cartridges 50s Made _____ Cartridges 100s Made _____ Number Vials Made _____

Date Certified _____ Certificate Number _____

Original Expiration Date _____ New Expiration Date _____

Quantity Certified: Number _____ Cartridge/50 _____

Number _____ Cartridge/100 _____

Amended by FDA _____ Amended Expiration Date _____

Amended Quantity _____ Cartridges/50 _____

_____ Cartridges/100 _____

DATE AND DISPOSITION MADE	On Hand This Date: C/50	C/100

REQUEST FOR ASSAY

Diska DATE_____

Item	Lot No.	Assay Type Required		K.B. ASSAY	REASSAY
		FDA	CANADA		

Date Completed_____

By_____

Remarks:

PRODUCT NAME AND NUMBER _____

AFFIX BELOW A FINISHED, PROPERLY CODED LABEL OF THE PRODUCT INDICATED ON THIS FORM SHOWING LOT NUMBER AND EXPIRATION DATE.

DATE	AFFIX LABEL HERE	AFFIXED BY	AMOUNT CTGS. LABELLED	LABELLED BY	TOTAL LABELS USED	INVENTORY	

Date of
Batch Release

Original Quantity
50 100

Description
Location

Stock No.

Date	Jiffy	50	100	Date	Jiffy	50	100	Date	Jiffy	50	100

Batch No. Symbol Exp. Date Cert. Date Cert. No.

Pfizer

SECTION 3r

The name of each person responsible for each of the above operations and information concerning his scientific training and experience:

A. ANTIBIOTIC ELUTION SUSCEPTIBILITY TESTING DISKS

T. J. McBride, Ph. D.
Technical Manager, Microbiology
Pfizer Diagnostics
199 Maywood Avenue
Maywood, New Jersey 07607

Education

B. A., Bacteriology	University of Kansas	1949
M. A., Bacteriology	University of Kansas	1950
Ph. D., Medical Microbiology	Northwestern University	1953

Experience

Pfizer Inc.

Section Leader, Bacteriology Antibiotic Research Department	1953-1961
Manager, Cancer Chemotherapy Screening and Evaluation Department	1961-1970
Technical Manager, Pfizer Diagnostics Microbiology Division	1970-Present

Publications

A total of 27 in fields of bacteriology, evaluation of new antibiotics, tissue culture and anti-tumor screening

---

Dennis A Rosenthal, B. S.
Production Supervisor, Disks
Pfizer Diagnostics
199 Maywood Avenue
Maywood, New Jersey 07607

Education

B. S., Biology	City College, New York	1971

Pfizer

Experience

Clinical Technologist Columbus Hospital, New York, New York	1971- 1972
Production Supervisor, Antibiotic Sensitivity Disks, Pfizer Diagnostics, Maywood, New Jersey	1972- Present

---

Joseph L. Hackett, Ph. D.
Supervisor
Microbiology Quality Control
Pfizer Diagnostics
199 Maywood Avenue, Maywood, New Jersey 07607

Education

B. Sc., Medical Technology	Ohio State University	1959
M. Sc., Clinical Pathology	Ohio State University	1963
Ph. D., Clinical Pathology	Ohio State University	1968

Experience

Research Assistant Infectious Diseases Laboratory Ohio State University Hospital	1960- 1967
Quality Control Manager, Courtland Scientific Products Division, Abbott Laboratories	1967- 1969
Microbiology Section Head, Reference Laboratories North Hollywood, California	1969- 1972
Supervisor, Microbiology Quality Control Pfizer Diagnostics	1972- Present

Publications

Total of four in fields of infections diseases

G. D. Patel, M. S.
Quality Control
Disk Assays
Pfizer Diagnostics
199 Maywood Avenue
Maywood, New Jersey 07607

Pfizer

Education

B. S., Biology	B. A. College of Agriculture Anand, Gujarat State, India	1962
M.S., Food Microbiology	Texas A&M University	1964

Experience

Instructor, Microbiology and Plant Pathology B. A. College of Agriculture	1962- 1964
Research Assistant, Dairy Science Department Texas A&M University	1965- 1966
Research Assistant University of Chicago Medical School Billing's Hospital Clinical Microbiology	1966- 1967
Quality Control Pfizer Diagnostics	1967- Present

Richard B. Dardas, Ph. D.
Manager, Microbiology Quality Control
Pfizer Diagnostics
199 Maywood Avenue, Maywood, New Jersey 07607

Education

B. S., Biology	Albion College	1957
M. S., Microbiology	Michigan State University	1959
Ph. D., Immunochemistry	Michigan State University	1963

Experience

Research Assistant Michigan State University	1957-1959
Research Fellow, Michigan State University	1959-1963
Staff Immunologist, Pfizer	1963-1967
Supervisor Chemical Res. & Dev., Pfizer	1967-1970
Supervisor Quality Control, Pfizer	1970-1972
Manager Quality Control Microbiology, Pfizer	1972-Presen

Publications

Two in the field of microbiology

Pfizer

SECTION 1

A full list of the articles used as components of the drug. This list should include all substances used in the fermentation, synthesis, extraction, purification or other method of preparation of any antibiotic and in the preparation of the finished dosage form, regardless of whether they undergo any change or are removed in the process. Each substance should be identified by its established name, if any, or complete chemical name, using structural formulas when necessary for specific identification. If any proprietary preparation is used as a component, the proprietary name should be followed by a complete quantitative statement of composition. Reasonable alternatives for any listed substance may be specified.

[redacted]

Pfizer

SECTION 7

A full statement of the composition of the drug. The statement shall set forth the name and amount of each ingredient, whether active or not, contained in stated quantity of the drug in the form in which it is to be distributed, as for example, amount per tablet or per milliliter, and a batch formula representative of that to be employed for the manufacture of the finished dosage form. All components should be included in the batch formula regardless of whether they appear in the finished product. Any calculated excess of an ingredient over the label declaration should be designated as such and per cent excess shown. Reasonable variations may be specified.

[redacted]

Pfizer

SECTION 3

A complete description of the methods and processes used in manufacturing, packing and labelling of the drug to preserve its identity, strength, quality and purity in conformity with good manufacturing practices including:

Pfizer

SECTION 3a

Name and location of each plant conducting the operations

A. EUGONIC BROTH AND BUFFERED SALINE

Eugonic Broth and Buffered Saline are manufactured, labelled and controlled at our plant at 199 Maywood Avenue, Maywood, New Jersey 07607. This plant is registered with the FDA as a diagnostic manufacturing facility on Form FD2656 (June, 1973).

Pfizer

SECTION 3b

Whether or not each lot of raw materials is given a serial number to identify it, and the use made of such numbers in subsequent plant operations

A. EUGONIC BROTH AND BUFFERED SALINE

Invoice identity, quantity and condition of containers are checked by receiving department personnel and the material is delivered to a designated quarantine area.

Quality Control is notified of arrival, location and invoice conformity.

An identification number is assigned to each container to facilitate record maintenance relating specific raw materials to each finished product. This information appears on individual prepared medium batch records.

A seven digit raw material number is assigned to each raw material lot which distinguishes it from any other lot of raw materials.

Pfizer

SECTION 3c

Precautions to assure proper identity, strength, quality and purity of the raw materials, whether active or not, including the specifications for acceptance and methods of testing for each lot of raw material used in the fermentation, synthesis, extraction, and purification of the drug and for each ingredient used in the manufacture of the drug that is to be dispensed.

A. EUGONIC BROTH AND BUFFERED SALINE

The raw materials are sampled according to an approved sampling plan as soon as feasible after receipt. Sampling operations include:

1. Specification of label identity

2. Inspection of container condition

3. Congruity of appearance with label identity

4. Presence of extraneous matter or other deficiency of quality

The raw material sample is tested by Quality Control according to the testing pattern indicated on the raw material specification following required test procedures.

Based on a review of test protocols by qualified personnel, materials are approved as meeting purchasing specifications. Approval is made in writing for use by authorized personnel as a raw material release document.

Containers are labelled to indicate description, inventory number, raw material lot number, release date, and approved signature and are then released for manufacturing processes. (See release raw material stickers and disposition certificates in Section 3q.)

Pfizer

SECTION 3d

If it is a drug produced by fermentation

Does not apply.

Pfizer

SECTION 3e

If it is a drug that is synthesized by chemical processes, a detailed description of each chemical reaction with graphic formulas used to produce the drug, including the names and amounts of all substances used in the process.

Does not apply.

Pfizer

SECTION 3F

Method of preparation of the master formula records and individual batch records and manner in which these records are used

A. EUGONIC BROTH AND BUFFERED SALINE

The Manufacturing Instruction Brief serves as the master formula record and includes all materials used, the nature of those materials including product codes, amount of materials, steps in the manufacturing process, equipment used, conditions of the process, a brief packaging and container description, and Quality Control tests. Individual medium batch records are derived from the master formula record in the Manufacturing Instruction Brief.

This information is used in the preparation of each batch of medium and buffered saline and is filed for a period of one year after the expiration date of the product or two years after final distribution, whichever is longer.

Pfizer

SECTION 3G

Number of individuals checking weight or volume of each individual ingredient entering into each batch of the drug.

A. EUGONIC BROTH AND BUFFERED SALINE

A responsible individual reviews the individual medium batch records, weighs and measures the ingredients for the finished product. The weight and volume are signed by the weigher and by a checker who confirms the weight, volume and addition of the material as noted on the individual medium batch record.

Pfizer

SECTION 3h

Whether or not the total weight or volume of each batch is determined at any stage of the manufacturing process subsequent to making up the batch according to the formula card, and at what stage and by whom this is done

A. EUGONIC BROTH AND BUFFERED SALINE

After medium or saline ingredients have been dissolved in water, and before the filling process, the volume of the product is confirmed by two qualified individuals. Visible characteristics are confirmed prior to filling.

SECTION 31

At what point in the process the drug is mixed homogeneously and a description of the equipment used for this purpose and its total capacity in terms of pounds, kilograms, gallons or liters of the drug and the maximum quantity of the drug that is mixed in such equipment.

A. EUGONIC BROTH AND BUFFERED SALINE

After the ingredients are weighed, they are added to a measured volume of distilled water that may contain up to as maximum process capacity.

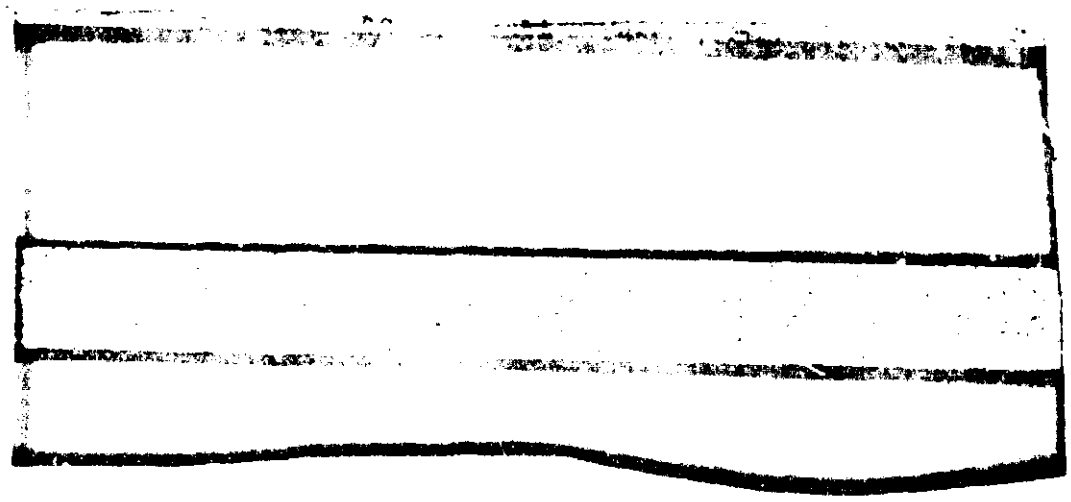
Mixing may be performed with a "Lightning" type of agitator or its equivalent to dissolve ingredients.

Pfizer

SECTION 3j

A description, where applicable, of all equipment used in the fermentation, synthesis, extraction, purification, filtration, sterilizing, grinding, blending, mixing, tableting, encapsulating, filling, packaging and labelling of the drug

A. EUGONIC BROTH AND BUFFERED SALINE



Pfizer

SECTION 3R

If it is a sterile drug, a description of the methods used to insure the sterility of each batch and the controls used for the maintaining of its sterility, including a detailed description of the sterile areas where the drug is produced and packaged

A. EUGONIC BROTH AND BUFFERED SALINE

[redacted]

Pfizer

SECTION 31

Additional procedures employed which are designed to exclude contaminants (e.g., other drug substances, extraneous materials, etc.) and otherwise assure proper control of the product

A. EUGONIC BROTH AND BUFFERED SALINE

Preparation and filling are processed separately and scheduled to avoid contamination of medium and saline with other products being simultaneously manufactured.

Vessels, mixers and filling equipment are thoroughly cleaned after each production run and inspected before a new manufacturing process is initiated.

Facilities for preparation of these products are inspected before each manufacturing process.

Finished products are sampled by Quality Control and tested for identity and absence of extraneous matter.

Pfizer

SECTION 3m

Adequate information with respect to the characteristics of and the test methods employed for the container, closure, or other component parts of the drug container to insure their suitability for the intended use.

A. EUGONIC BROTH AND BUFFERED SALINE

Glass test tubes are checked for breaks, clarity, size and fit with the Autobac 1 cuvette. The torque of the closure is measured for adequacy.

Pfizer

SECTION 3n

Controls used in the packaging and labelling of each batch to insure the standards of identity, strength, quality and purity of the drug

A. EUGONIC BROTH AND BUFFERED SALINE

The correct number of labels are released against the Manufacturing Instructions Brief by Quality Control and are checked out of a label room and verified by two responsible individuals who confirm the product number and description.

Lot numbers are verified upon their printing on labels by the operator and an additional qualified person.

A designated individual determines if labels are fixed in the proper position, if information is clearly visible and if adherence to surfaces is adequate.

All unused labels in a production run are counted and a tally is entered into the label accountability book. Unused labels are destroyed.

Pfizer

SECTION 30

Precautions to check the total number of finished packages produced from a batch of the drug with the theoretical yield

A. EUGONIC BROTH AND BUFFERED SALINE

A designated individual checks the number of finished packages for the total number of containers and compares this total with the theoretical yield noted in the individual medium batch records. Explanation of variance from theoretical yield is logged.

Pfizer

SECTION 3p

Precautions, to insure that each lot of the drug is packaged with the proper label and labelling, including provisions for labelling, storage and inventory control

A. EUGONIC BROTH AND BUFFERED SALINE

Labels on finished goods are checked by Quality Control for identity, lot number and correctness and the yield statement is then reconciled with the label accountability log.

The total number of labels used are recorded and excess labels in the labelling area are removed before the next operation commences. Labels are fixed to the individual batch records of each lot of finished produc..

(Pfizer)

SECTION 3q

Copies of all printed forms used by the applicant in the manufacture, packaging and labelling of a batch

A. EUGONIC BROTH AND BUFFERED SALINE

See attached.

PACT NO. PREPARED MEDIA BATCH CONTROL RECORD LOT NO.

PFIZER DIAGNOSTICS
RAW MATERIAL

Description: _____

Inventory No._____

R.M. Lot No._____

Release Date:_____

Approved Signature_____

PFIZER DIAGNOSTICS
RAW MATERIAL

Description:_____

Inventory No._____

R.M. Lot No._____

Release Date:_____

Approved Signature_____

DRY POWDER MEDIA PRODUCTION AND CONTROL RECORD

CONTROL NUMBER _____ MANUFACTURING DATE _____

Pfizer

SECTION 3r

The name of each person responsible for each of the above operations and information concerning his scientific training and experience

A. EUGONIC BROTH AND BUFFERED SALINE

Edward J. Muller, Jr.
Production Manager
Pfizer Diagnostics
199 Maywood Avenue
Maywood, New Jersey 07607

Education

B. S., Chemistry	Missouri State College	1966

Experience

Control Chemist	Lever Bros. Edgewater, N. J.	1965- 1966
Research Chemist	Interchemical Co. Carlstadt, N. J.	1966- 1968
Pfizer Inc., Parsippany, N. J.		
Quality Control Supervisor		1968- 1969
Production Supervisor		1969- 1971
Processing Manager		1971- 1972
Pfizer Inc., Maywood, N. J.		
Production Manager		1972- Present

(Pfizer)

Glenn Abello
Production Supervisor
Pfizer Diagnostics
199 Maywood Avenue.
Maywood, New Jersey 07607

Education

B.S., Industrial Engineering	Northeastern University	1967
M.B.A., Business Administration	Columbia University	1972

Experience

Production Supervisor in the manufacture of Corning Ware - finishing and ceramming operations - Corning Glass Works	1972-1973
Production Supervisor Prepared Media Pfizer Diagnostics	1973-Present

---

Joseph L. Hackett, Ph. D.
Supervisor
Microbiology Quality Control
Pfizer Diagnostics
199 Maywood Avenue, Maywood, New Jersey 07607

Education

B. Sc., Medical Technology	Ohio State University	1959
M. Sc., Clinical Pathology	Ohio State University	1963
Ph. D., Clinical Pathology	Ohio State University	1968

Experience

Research Assistant Infectious Diseases Laboratory Ohio State University Hospital	1960-1967
Quality Control Manager, Courtland Scientific Products Division, Abbott Laboratories	1967-1969
Microbiology Section Head, Reference Laboratories North Hollywood, California	1969-1972
Supervisor, Microbiology Quality Control Pfizer Diagnostics	1972-Present

Publications

Total of four in fields of infections diseases

Pfizer)

Section 3r (continued)

G. D. Patel, M. S.
Quality Control
Disk Assays
Pfizer Diagnostics
109 Maywood Avenue
Maywood, New Jersey 07607

Education

B. S., Biology	B. A. College of Agriculture Anand, Gujarat State, India	1962
M.S., Food Microbiology	Texas A&M University	1964

Experience

Instructor, Microbiology and Plant Pathology B. A. College of Agriculture	1962-1964
Research Assistant, Dairy Science Department Texas A&M University	1965-1966
Research Assistant University of Chicago Medical School Billing's Hospital Clinical Microbiology	1966-1967
Quality Control Pfizer Diagnostics	1967-Present

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Pfizer

Section 7r (continued)

Richard B. Dardas, Ph. D.
Manager, Microbiology Quality Control
Pfizer Diagnostics
199 Maywood Avenue, Maywood, New Jersey 07607

Education

B. S., Biology	Albion College	1957
M. S., Microbiology	Michigan State University	1959
Ph. D., Immunochemistry	Michigan State University	1963

Experience

Research Assistant Michigan State University	1957-1959
Research Fellow, Michigan State University	1959-1963
Staff Immunologist, Pfizer	1963-1967
Supervisor Chemical Res. & Dev., Pfizer	1967-1970
Supervisor Quality Control, Pfizer	1970-1972
Manager Quality Control Microbiology, Pfizer	1972-Present

Publications

Two in the field of microbiology

Autobac 1

8. STABILITY INFORMATION

8. STABILITY INFORMATION

In addition to stability information, this section provides all the information which customarily was supplied in Sections 4 and 5 of the Form 5. It therefore contains other related information such as a complete description of the tests and methods of assay and other controls used during manufacture of the batch and after it is packaged.

A distinction is made between currently certifiable and non-certifiable items. The format of Sections 4 and 5 of the previously used Form 5 has been adhered to in order to facilitate review.

SECTION 4

A complete description of the tests and methods of assay and other controls used during the manufacture of the batch and after it is packaged.

Studies on Elution Disks for the Autobac I

Instrument for Antimicrobial Susceptibility Testing

General Background

The Autobac I is an automated device for the determination of the susceptibility of bacteria to therapeutic antimicrobial agents. The essential components are:

1) A buffered saline solution for the preparation of a standardized inoculum of the bacteria under study.
2) A premeasured volume of sterile Eugonic broth to support bacterial growth.
3) A specially designed multi-chambered cell (cuvette) which provides one reference chamber for uninhibited normal bacterial growth and twelve test chambers in each of which antimicrobial challenge of the bacteria takes place.
4) Paper disks containing antimicrobial agents. Such disks, designed to provide the appropriate amount of antimicrobial agent to each cuvette test chamber, are termed "elution disks".
5) An incubator/shaker in which inoculated cuvettes are incubated and agitated.
6) A special photometer and calculator for measuring uninhibited baseline growth and reporting any inhibition of bacterial growth in each of the twelve antimicrobial test chambers.

A pure culture of the organism under test is selected from the culture plate and suspended in buffered saline to within a standard light scattering range, and a 2 ml. aliquot of this suspension is added to a tube containing 18 ml. of sterile Eugonic broth. The inoculated broth is added to the cuvette and equally distributed among the thirteen chambers. Different antimicrobial disks are added to the twelve testing chambers and the cuvette

shaken and incubated for three hours to elute antimicrobial agents from the disks and maintain enumerating bacterial cells in suspension. The inhibition of growth by the antimicrobial agent is then measured in the Autobac I photometer system. (A more complete description of the operation of the Autobac I system can be found in Sections 7 (Use Manual), and 13 (Package Insert).

Among the factors critical in obtaining reproducible and proper functioning of this device is the ability to provide a reproducible appropriate amount of antimicrobial agent to each test chamber of the cuvette. This report discusses the characteristics and requirements of satisfactory elution disks. The term elution disks distinguishes disks devised for this system from the diffusion disks used in the Kirby-Bauer and other techniques for determining antimicrobial susceptibility on agar plates by diffusion of inhibitory concentrations of agent into the solid growth-supporting medium.

Summary

1. Disks manufactured by procedures described in this application dispense reproducible and predictable amounts of antimicrobial agents into the Eugonic broth as it is used in the Autobac I system.
2. For each antimicrobial agent impregnated in the paper disk, the ratio of the amount released by elution to that assayed by diffusion assay is constant.
3. These elution/diffusion ratios are not the same for all antimicrobial agents. In no case does the amount eluted exceed that applied to the disk, although it has been found to be less.
4. These ratios and the amount of antimicrobial agent eluted are not affected by disk age.
5. Except for Polymyxin B, elution of antimicrobial agents from disks attains maximal levels within three hours, frequently within ten minutes. Further elution removes little or no additional antibiotic.
6. Except for Penicillin G, Cephalothin, Erythromycin and Gentamicin, the eluted antimicrobial agent appears stable in solution for at least three hours.
7. The amount eluted and the elution rates are increased by shaking.
8. In our hands the precision of disk assay by microbiological diffusion methods is superior to those of disk assay by microbiological elution methods. Microbiological diffusion assays such as those described in CFR 147.1 for diffusion disks are appropriate methods of assaying certifiable elution disks designed for use in the Autobac 1 instrument.
9. For selected antimicrobial agents, e.g. Nitrofurantoin, Nalidixic Acid, ultraviolet absorption assay methods offer high precision.
10. A list of disks, potencies and ranges for which approval is being sought at this time is given in Table 9.

Introduction

Basic Information:

Information is presented on the following aspects of elution disks, largely in tabular form:

1) Rates and amounts of elution of antimicrobial agents from various disks (Tables 1a-1v, 2).
2) Reproducibility of elution and diffusion assays (Table 3).
3) A summary of assay methodology used in the elution and diffusion assays (Tables 4, 5).
4) The effect of disk age on elution (Table 6).
5) The effect of elution conditions on antimicrobial agent elution (Tables 7, 8).
6) A summary of optimal disk masses and optimal mass ranges for all elution disks studied to date (Table 9).
7) A list of disks for which approval is being sought at this time (Table 10).
8) A comparison of elution and diffusion disk potencies and expiration date claims (Table 11).
9) A report on elution studies including the effect of shaking, on Tetracycline and Doxycycline elution disks.
10) Reports describing diffusion assays for Penicillin G, 0.2 U. and Ampicillin, 0.25 mcg. disks.
11) A report describing studies on Nalidixic Acid and Nitrofurantoin elution disks.
12) Appendix 1, showing schematically the operation of Autobac 1.
13) Test procedures for non-certifiable elution disks.
14) Specifications and stability data for Eugonic Broth and Phosphate Buffered saline (Inoculum Standardization Solution).

General Considerations:

These studies on disks were initiated prior to the final establishment of appropriate disk potencies for the Autobac 1 system (Studies justifying the potencies selected are described in Section II. G and 16. vi-viii). For this reason, some assays were conducted on disks with potencies slightly different from those finally recognized as optimum. For example, the elution of Ampicillin was studied on disks of nominal potencies of 0.25, 5.0 and 7.5 mcg., although the potencies finally found to be optimal to ensure correlation between Autobac 1 and Kirby-Bauer data were 0.22 and 3.6 mcg for Gram positive and Gram negative microorganisms, respectively. Elution studies were conducted on Tetracycline disks of nominal potencies 0.5, 1.2, and 5 mcg., although the optimal potencies were eventually established to be 0.5 and 1.2 mcg. for Gram positive and Gram negative microorganisms, respectively. Limited studies were conducted also on standard diffusion disks with potencies that were frequently significantly different from those of elution disks. In no case have we observed a significant difference between the elution characteristics of high and low potency disks, and the data herein can therefore be assumed with full confidence to apply to disks of the potencies finally selected.

All certified disks were manufactured by the exact procedures described in documents on file with FDA describing the manufacture of diffusion disks, except that the concentration of the antimicrobial agent in the impregnating solvent was adjusted as necessary to yield the desired disk potency. These procedures have been in use since 1961 and have been demonstrated by data already on file to yield stable disks eminently satisfactory and useful in the agar diffusion method of Kirby-Bauer, et.al. for antimicrobial susceptibility testing. These manufacturing procedures are described in detail in Section 7*of this application.

The data herein applies only to disks manufactured by Pfizer Diagnostics and eluted with Eugonic broth-saline in the Autobac 1 cuvette and shaker. Disks suitable for diffusion assays but manufactured differently can give

* Section 7. III, Disposables = Sections 1-3 of the Form 5.

significantly different elution patterns. Details of eluant volume, shaking speed, etc., may also introduce differences in the elution behavior; e.g. disks in cuvettes that are not shaken give significantly slower elution than do disks in cuvettes which are shaken.

Nalidixic acid and Nitrofurantoin disks are manufactured in an entirely analagous manner. We have described herein elution data for Nalidixic Acid and Nitrofurantoin disks for information only. It is recognized that these are not subject to the licensing or certification requirements of antibiotic susceptibility disks; neither are the buffered saline or Eugonic broth.

Studies are also reported on certain certifiable disks for which approval is not being requested now. It is our intention to request certification of these at a later date.

Disks for which Certification is Requested:

In this application we are requesting approval only of the following fifteen disks for use in the Autobac 1 system. Data regarding other disks should, at this time, be considered as general substantiating information.

Ampicillin	3.6 mcg (for G- organisms)
Carbenicillin	120 mcg (for E. coli, Proteus sp. + Pseudomonas aeruginosa)
Cephalothin	15 mcg. (for G+ and G- organisms)
Chloramphenicol	4 mcg. (for G- organisms)
Clindamycin	2 mcg. (for G+ organisms)
Colistin	13 mcg. (for G- organisms)
Erythromycin	2.5 mcg. (for G+ organisms)
Gentamicin	9 mcg. (for G+ and G- organisms)
Kanamycin	22 mcg. (for G- organisms)
Methicillin	5 mcg. (for G+ organisms)
Penicillin	0.2U. (for G+ organisms)
Polymyxin B	12.5U. (for G- organisms)
Tetracycline	0.5 mcg. (for G+ organisms)
Tetracycline	1.2 mcg. (for G- organisms).
Vancomycin	30 mcg. (for G+ organisms)

Discussion

Elution Assays:

Elution assays were conducted by charging a standard Autobac 1, thirteen-chamber, plastic, cuvette with 20 ml of a mixture containing 18 ml of Eugonic broth and 2 ml of Phosphate Buffered saline, which exactly simulates the inoculated growth medium used in the normal operation of the Autobac 1 for antimicrobial susceptibility testing. The cuvette was manipulated to obtain an even distribution of the eluant in the thirteen cells, 1.54 ml per cell. Each of eight chambers were then charged with a disk of the antimicrobial agent under investigation. In an initial study, one cuvette was shaken and incubated for three minutes, others for 10, 20, and 30 minutes. In later studies cuvettes were prepared and incubated for ten, 30, 60, 90, and 180 minutes to more nearly simulate the actual incubation period recommended for susceptibility testing; the three minute elution was omitted since the initial study showed incomplete elution of several antimicrobial agents at that time. Immediately after shaking at 220 revolutions per minute in a 36°C. incubator specially designed for use with this system, the cuvette was removed and the eluate withdrawn separately from each chamber by pipette. The eluate was transferred within 30 minutes to stainless steel cups on microbiological assay plates prepared in accord with CFR methods for the antibiotic under study, (modified where necessary to accommodate the actual concentration expected in the eluate). In those cases where the eluate solutions were significantly diluted prior to assay, the CFR specified buffer was used as the diluent so that the standard cup and the assayed cup contained antibiotic in substantially the same medium. In those cases where the concentration of antibiotic in the eluate was low, the standards were

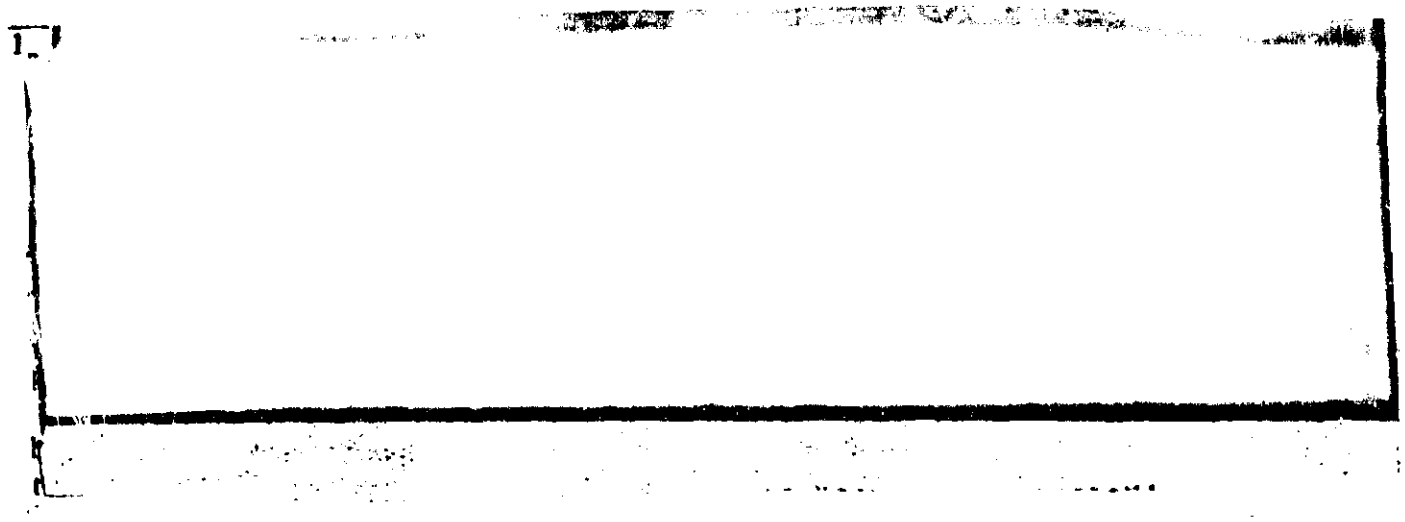
prepared in Eugonic broth-saline[1] to ensure that both standard and assayed solutions were analagous.

Following overnight incubation, zone sizes were measured and potencies in the eluate were determined by standard methods. The amount of antibiotic eluted from each disk was obtained by multiplying the concentration of antibiotic in the eluant solution by 1.54, the number of ml of eluant in each cuvette chamber. Details of the exact assay procedure used for each antimicrobial agent are shown in Table 4.

Antibiotic Disk Diffusion Assays:

Diffusion assays were conducted by the standard CFR method for the antibiotic, modified where necessary by adjusting the disk concentrations used to generate the standard curve. Twelve disks were assayed on each of two successive days for the diffusion assays. Minor modifications, described in the appended report from F. C. Keenan to Dr. E. M. Cohen, dated 1/31/73, were utilized for Ampicillin 0.25 meg. and Penicillin G 0.2 U. disks.[a] Details of the potencies used for all antimicrobial agents are shown in Table 5.

With the Polymyxin B 12.5 U. disk, insufficient antibiotic was eluted to permit an effective assay on the eluant solution. In order to assess Polymyxin B elution from these disks, disks were eluted as described above,



immediately removed from the solution, blotted dry and then assayed by a diffusion method. Minor difficulties were encountered due to irregular zones in the original assay procedure, but multiple zone diameter measurements in the circular areas permitted an assay of reasonable precision.

Non-certifiable Disk Assays:

Nalidixic Acid and Nitrofurantoin were assayed by elution with Eugonic broth for predetermined periods as described above, blotted dry and re-eluted for extended periods in normal saline solution which shows a lower background at the measuring wavelength. In this way it was established that one hour elution yielded about 1-3% more Nalidixic Acid extraction than did 30 minute elution. In both cases, the antimicrobial agents eluted from the disks showed an ultraviolet absorption curve from 400-220 nm identical to that of the pure compound, when corrected for a minor absorption peak at about 260 nm originating from the paper.

Results:

Tables Ia-Iv show for each antimicrobial agent the lot numbers studied, their manufactured nominal concentration and the results of both diffusion and elution assays at periods ranging from ten to 180 minutes. In general, for at least one lot, elution assays were conducted at two to four weeks following manufacture and again at nine to twelve months. Also recorded under each antimicrobial agent in Tables I is the average ratio of the elution assay to the diffusion assay. When calculating these ratios, the elution assay was chosen from those values representing the maximum elution, e.g. for Polymyxin B only the three hour assay was utilized, for Penicillin G, Cephalothin, Erythromycin and Gentamicin the three hour assay was not used since there was consistent evidence of degradation at that period; the average is based on assays conducted between ten and 90 minutes. In other instances, e.g. Novobiocin, the ten minute assay was omitted in calculating elution assays since the data indicates that elution was incomplete at

that time; the elution assay value was calculated as the average of the 30 to 180 minute values.

A summary of the ratio of the elution assay to diffusion assay ratio is shown for each antibiotic in Table 2. If one takes into allowance the very significant variance in the assays, the data summarized in Table 2 shows the ratio of elution assay to diffusion assay to be constant for each antimicrobial agent, e.g. the range for 7.5 mcg. Ampicillin disks was 0.70-0.92 with an average of $0.80 \pm 1\,SD = 0.08$. As nearly as the accuracy of microbiological assay allows, it can be concluded that the diffusion assay is an accurate predictor of the amount of antimicrobial agent that can be eluted from a disk. The precision of assays by both methods is shown in Table 3. In few cases the elution assay was more reproducible than the diffusion assay. Consideration of the fact that the diffusion assay of disks is well established in both our laboratories and yours, of the small amounts of antibiotic involved, and of the higher precision of this assay leads to its nomination as the method of choice at this time for certifiable antibiotics. For Nalidixic Acid and Nitrofurantoin, the assay method of choice is elution followed by an ultraviolet spectrophotometric assay. The reproducibility of this assay, which is analagous to USP methodology, is higher than that of microbiological assays. It is entirely likely that analagous, more precise procedures can be developed for antibiotics showing specific ultraviolet absorption at appropriate wavelengths, though none are offered at this time.

The data in Table 2, which summarizes data from Tables 1a-1v, shows that the ratio of elution to diffusion assays, though constant for each antimicrobial agent, differs significantly from one another. This should not be unexpected for the following reason. For the small concentrations of antimicrobial agent involved, it can reasonably be anticipated that there will be some significant amount of irreversible absorption onto the paper of the disk. It can also be anticipated that the extent of such irreversibly bound antimicrobial agent will not necessarily be the same for the different antimicrobial agents. In conducting diffusion assays the manufactured disk is compared

to a series of standard disks from which the exact extent of antimicrobial agent transfer to the agar plate is not known. The objective of the assay is not to determine the amount of total antimicrobial agent actually in the test disk, but the relationship to the amount transferred from the test disk into agar to that transferred from the standard disk into agar. An accurate assay is therefore obtainable if the amount of antimicrobial agent actually diffusing from the disk corresponds to the amount applied to the standard disk or if the amount which diffuses is reduced by irreversible absorption. An accurate assay requires only that irreversible absorption be constant for the standard disks of that antimicrobial agent. In elution assays, on the other hand, antimicrobial agent is eluted from the paper into solution. This solution is then compared directly to standard solutions. Irreversible absorption by the disk will reduce the amount eluted and differences in elution of different agents from paper will be demonstrable by this technique. It is reassuring that in no case did the amount of antimicrobial agent eluted from the paper significantly exceed that calculated to be applied. (Our normal practice in manufacturing elution disks of a given nominal potency for this study was to impregnate approximate a 25% overage. If elution from the paper was complete, then one should expect to find 125% of the nominal value by elution.)

Again, it must be emphasized that the limited precision of both the diffusion and elution assays, with coefficients of variation averaging 15% for the diffusion assays and 19% for the elution assays, must be taken into account in interpreting this data. If, for example, the "true value" of the diffusion assay is 10 mcg., with a coefficient of variation of 15%, and the "true value" of the elution assay is 7.5 mcg., with a coefficient of variation of 19%, the "true ratio" would be 7.5/10=0.75. However, a difference of only one standard deviation in each assay could lead to ratios of 6.1/1.15=0.53, or of 8.9/8.5=1.05. Also relevant is the wide range of disk concentrations, typically 68-150% of label, which are tolerable without significant decrease in the correlations between bacterial susceptibility as determined by the Kirby-Bauer methods and the Autobac 1 procedure, and the invariance of the amount of antimicrobial agent diffusable, hence potentially elutable, from disks, as shown by stability studies on diffusion disks, data already on file with you.

Effects of Disk Age on Elution:

The effect of disk age on the amount of antimicrobial agent eluted was also studied. In addition to examining elution disks prepared especially for the Autobac I instrument, supplementary data was attained by studying elution from standard certified disks manufactured for use in diffusion assays. Data on file in support of our assigned expiration date for diffusion disks establishes that there is no significant loss in antibiotic diffusable from a disk over the life of the disk to expiration. The data in Table 6 shows that for all disks studied, the elution to diffusion ratios remain constant, considering the precision of the assay methodology, for periods approximating one year. It seems highly improbable that ratios would change during the established expiration date.

Theoretically, irreversible absorption of antimicrobial agent to the impregnated paper must require time. In practice, the time required to attain equilibrium absorption seems short, much shorter than our usual four week manufacture and certification cycle. In one study Novobiocin disks containing 2.5 mcg. of Novobiocin were accurately prepared by hand, and eluted 18 hours later into a buffered normal saline, pH 7, of the same osmolarity as Eugonic broth utilized in the Autobac I system. Elution of Novobiocin for 30 minutes yielded 2.16 mcg. per disk, 87% of the amount applied, while a disk manufactured 90 days earlier in an identical fashion showed an elution of 2.12 mcg., 85% of the amount applied. A 10 mcg. Novobiocin disk manufactured in our normal manner three months before assay yielded 81% of the applied antibiotic after 30 minutes elution. These three results are not significantly different, and indicate that for Novobiocin "absorption equilibrium" was attained in less than 18 hours. An ultraviolet spectrophotometric assay was used in this study which is not included in Table 1p. Vancomycin showed results identical, in a similar study, to those recorded for Novobiocin. In another separate study, described in detail in Appendix 2, "Measurement of Rates of Elution of Labeled Tetracyclines from Paper Sensitivity Discs into Eugonic Broth Medium," Joseph F. Dooley, October 25, 1972, it was demonstrated that Doxycycline and Tetracycline are substantially completely

eluted from hand-made disks prepared a few days earlier. This study utilized tritium labeled antibiotics and a radioactive assay methodology. The less precise microbiological assay methodology used to generate the data on Doxycycline and Tetracycline in Table 6 shows elution to diffusion ratios relatively constant over periods up to two years, albeit slightly lower than those found by the radioactive studies on disks a few days old.

In view of the fact that our normal manufacturing and FDA certification cycle requires a minimum of three to four weeks between impregnation and release, there appears to be virtual certainty that any disk manufactured would have reached "absorption equilibrium" prior to use.

Effect of Rotary Shaking on Elution Rates in the Autobac 1 System:
Several studies on the effects of shaking on elution rates were conducted. The most detailed titled "Measurement of Rates of Elution of Labeled Tetracyclines from Paper Densitivity Discs into Eugonic Broth Medium," Joseph F. Dooley, October 25, 1972, is presented in Appendix 2. It is clear from the data recorded therein that shaking significantly increases the rates of elution of Doxycycline and of Tetracycline from 1.2 mcg. Tetracycline disks and 2 mcg. Doxycycline disks.

Similar studies on Nitrofurantoin 25 mcg. and Nalidixic Acid 15 mcg. disks showed qualitatively similar results. The exact findings for Nalidixic Acid are detailed in Table 7. A phosphate buffered saline of the same pH and osmolarity as Eugonic broth was used for this study, in conjunction with an ultraviolet spectrophotometric absorption assay method. (Other studies on higher concentration Nalidixic Acid disks had shown identical elution by Eugonic broth and by this phosphate buffered saline at 30 minutes. Saline, without ultraviolet absorption background, is preferable for quantitative studies of this type.)

In another study, conducted with Nalidixic Acid disks and phosphate buffered saline eluant, the effect of the number of disks on elution was

examined. Details are shown in Table 8. It is clear that increasing the number of disks from one per chamber to two or three per chamber decreases the elution rate of antibacterial agent from the disks. This is unlikely to be a saturation phenomenon since the solubility of Nalidixic Acid in pH 7 buffer is higher than the actual concentrations attained in these studies. All disks, whether added one, two or three per cuvette chamber, were thoroughly wetted by the eluant in this study. It appears that the elution rate of antibacterial agent from the interior of a stack of three disks is significantly slower than is the elution rate from a single disk, even with shaking.

Comparison of Pfizer Diagnostics and Other Manufacturer's Disks:

Limited studies in the Autobac Incubator/Shaker compared elution of antibiotic from ten of our disks with comparable disks manufactured by Difco Laboratories and Baltimore Biological Laboratories. These studies were conducted with commercially available diffusion disks. Their potencies, therefore, do not necessarily correspond to those of elution disks manufactured for use with the Pfizer Autobac I system. On balance, disks manufactured by Pfizer and by Difco Laboratories showed elution of substantially the same amount of antimicrobial agent. The BBL disks, in contrast, showed significantly higher amounts of elutable antimicrobial agent. It appears that not all manufacturer's disks with approximately the same diffusion assay will yield the same amount of antibiotic on elution in the Autobac I system.

Optimum Disk Potencies and Acceptable Ranges:

Table 9, which is based in its entirety on work described in Section II. G. of this application, summarizes the final nominal disk potencies most suitable for elution disks manufactured by our methodology as intended for use with the Autobac I system. The final column of this table shows the potency range which can be tolerated while still maintaining the same high interpretative correlation between antibiotic susceptibility results as determined by the Autobac I and Kirby-Bauer procedures. It should be noted that the % optimum potency range equals or exceeds that now assigned to certifiable diffusion disks for 16 of the 26 disks studied. The remainder have % optimum ranges from 80-150% of label (e.g., Chloramphenicol, Carbenicillin, Kanamycin) to 80-125% [e.g., for Ampicillin (3.6 meg disk) and Nitrofurantoin].

The disks for which approval is sought at this time, with permissible ranges, are listed separately in Table 10.

Assay Procedures:

For antibiotic disks with % optimum potency ranges of 80-150% or broader, the standard two day CFR assay, modified by appropriate upward or downward adjustment of the standard curve, appears to offer adequate accuracy for certification and control procedures. For those disks with narrower ranges, e.g. Ampicillin 3.6 mcg. (80-125%) and Tetracycline 1.2 mcg (80-130%), obtaining adequate accuracy with the CFR assay may require more replication, e.g. four replicate assays rather than the two required by the present CFR method.

For Nitrofurantoin 15 mcg. (80-125%) and Nalidixic Acid 15 mcg. (68-150%) the elution and ultraviolet absorption assay described herein offers adequate accuracy. Initial studies suggest that ultraviolet absorption assays will offer higher precision for selected antibiotic disks as well. Details of these are not available at this time, since the studies are currently not yet completed. Experiments performed for the purpose of determining the required test pattern required for increased precision have been performed with Ampicillin and Doxycycline. Data derived from these experiments is presented in Table 3a. The requisite number of standard curves, test plates and disks required to provide assays of appropriate precision is listed in Table 3b.

Elution Disk Stability and Expiration Dates:

For 15* of the certifiable elution disks intended for use with the Autobac 1 system, diffusion disks of substantially equal or lower nominal potency than the corresponding elution disk are currently manufactured for use in agar diffusion susceptibility testing. For these 15 elution disks*, we claim an expiration date equal to that established for the diffusion disks. (Discussions with Dr. W. Wright, Messrs. G. Carter and R. Norton in November, 1972, led to informal approval of this claim. Chloramphenicol 4 mcg. and *Ampicillin (3.6 mcg), Bacitracin (18 U), Carbenicillin (120 mcg), Chloramphenicol (4 mcg), Clindamycin (2 mcg), Colistin (13 mcg), Erythromycin (2.5 mcg), Gentamicin (9 mcg), Kanamycin (22 mcg), Lincomycin (2.4 mcg), Methicillin (5 mcg), Neomycin (24 mcg), Oleandomycin (6 mcg), Streptomycin (20 mcg), and Vancomycin (10 mcg).

Autobac 1 system.

a) Specifications for Eugonic broth for the Autobac 1.

b) Stability data for Eugonic broth.

c) Specifications for Inoculum Standardization Solution for the Autobac 1.

d) Stability data for Phosphate Buffered saline (Inoculum Standardization Solution).

e) Test procedures for Nalidixic Acid and Nitrofurantoin disks.

TABLE 1a - Part 1

Ampicillin

Lot Numbers	2266	2266	2421	2422
Manufactured	4/72	4/72	12/72	12/72
Mfld. Nominal Conc.* mcg.	7.5	7.5	7.5	7.5
Age at Assay	2 wks.	8 mos.	1 mo.	1 mo.
Diffusion Assay	8.4	8.7	8.0	7.9
Elution Assay, 10 min.	7.4	5.9	6.2	6.3
20 min.	7.9	---	---	---
30 min.	7.7	6.2	7.0	6.6
60 min.	---	6.3	6.5	6.2
90 min.	---	6.5	6.3	5.8
180 min.	---	5.7	5.9	5.1
Average Elution Assay (all times)	7.7	6.1	6.6	6.0
Ratio of Elution to Diffusion Assay	0.92	0.70	0.83	0.76

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE Ia - Part 2

Ampicillin

Lot Numbers	2530	2660	2736
Manufactured	2/73	5/73	8/73
Mfrd. Nominal Conc.*, mcg.	5	5	5
Age at Assay	6 mos.	4 mos.	1 mo.
Diffusion Assay	6.7	6.1	6.0
Elution Assay, 10 min.	4.8	2.9	4.4
30 min.	5.5	4.4	4.3
60 min.	5.8	3.9	4.2
90 min.	4.6	4.2	4.9
180 min.	4.8	4.4	4.7
Average Elution Assay (all times)	5.1	4.0	4.5
Ratio of Elution to Diffusion Assay	0.76	0.66	0.75

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1a - Part 3

Ampicillin

Lot Numbers	2354	2459	2465
Manufactured	8/72	12/72	12/72
Mfd Nominal Conc.*, mcg.	0.25	0.25	0.25
Age at Assay	7 mos.	3 mos.	3 mos.
Diffusion Assay	0.18	0.29	0.27
Elution Assay, 10 min.	0.17	0.23	0.24
20 min.	---	---	---
30 min.	0.16	0.23	0.23
60 min.	0.16	0.23	0.24
90 min.	0.16	0.23	0.24
180 min.	0.16	0.22	0.23
Average Elution Assay (all times)	0.16	0.23	0.24
Ratio of Elution to Diffusion Assay	0.89	0.79	0.88

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1b - Part 1

Bacitracin

Lot Numbers	2239	2239	2445	2446
Manufactured	3/72	3/72	12/72	12/72
Mnfd. Nominal Conc.*, U.	12	12	12	12
Age at Assay	4 wks.	13 mos.	4 mos.	4 mos.
Diffusion Assay	12.4	12.4	14.0	13.2
Elution Assay, 10 min.	10.4	13.4	18.3	16.2
20 min.	10.9	---	---	
30 min.	10.9	14.2	18.2	16.0
60 min.	---	15.2	18.5	18.3
90 min.	---	14.8	18.8	16.0
180 min.	---	15.6	18.2	15.9
Average Elution Assay (all times)	10.7	14.6	18.4	16.3
Ratio of Elution to Diffusion Assay	0.86	1.17	1.31	1.25

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1b - Part 2

Bacitracin

Lot Number:	2703	2704	2705
Manufactured	7/73	7/73	7/73
Mfd. Nominal Conc.*, U.	18	18	18
Age at Assay	2 mos.	2 mos.	2 mos.
Diffusion Assay	20.3	19.2	20.2
Elution Assay, 10 min.	25.6	25.6	23.6
30 min.	24.1	26.0	23.3
60 min.	27.9	26.6	25.3
90 min.	25.6	24.1	25.2
180 min.	25.4	22.2	25.1
Average Elution Assay (all times)	25.7	24.9	23.4
Ratio of Elution to Diffusion Assay	1.27	1.30	1.22

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE Ic

Carbenicillin

Lot Numbers	2291	2291	2531	2577
Manufactured	5/72	5/72	2/73	4/73
Mnfd. Nominal Conc.*, mcg.	120	120	120	120
Age at Assay	2 wks.	12 mos.	3 mos.	2 mos.
Diffusion Assay	144	117	142	132
Elution Assay, 10 min.	87	84	90	91
20 min.	87	---	---	---
30 min.	88	89	96	89
60 min.	---	89	97	90
90 min.	---	90	97	90
180 min.	---	90	98	95
Average Elution Assay (all times)	87	88	96	91
Ratio of Elution to Diffusion Assay	0.61	0.75	0.68	0.67

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE Id

Cephalothin

Lot Numbers	2271	2271	2423	2424	2532
Manufactured	4/72	4/72	12/72	12/72	2/73
Mufd. Nominal Conc.*, mcg.	15	15	15	15	15
Age at Assay	2 wks.	9 mos.	1 mo.	1 mo.	7 mos
Diffusion Assay	17.7	17.3	18.9	18.1	19.4
Elution Assay, 10 min.	17.6	18.4	18.3	20.2	14.9
20 min.	20.3	---	---	---	---
30 min.	18.5	15.7	17.3	16.9	15.3
60 min.	---	14.6	17.6	16.7	14.6
90 min.	---	18.8	18.6	17.7	15.0
180 min.	---	13.5	17.6	14.7	13.6
Average Elution Assay (10-90 min. values)	18.8	16.9	18.0	17.9	15.0
Ratio of Elution to Diffusion Assay	1.06	.98**	.95	.99	.77

*Impregnated product on disks were targeted to be 125% of this nominal potency.

**A third assay on lot 2271 @ 17 mos. yielded a diffusion assay of 17.2 mcg., elution assay of 14.3; elution to diffusion ratio = 0.83

TABLE 1e

Chloramphenicol

Lot Numbers	2253	2253	2015	2457
Manufactured	3/72	3/72	2/71	12/72
Mfrd. Nominal Conc.*, mcg.	7.5	7.5	5	5
Age at Assay	8 wks.	3 mos.	23 mos.	9 mos.
Diffusion Assay	11.5	10.8	6.8	5.8
Elution Assay, 10 min.	11.4	10.7	8.7	4.1
20 min.	14.8	---	---	---
30 min.	12.7	12.5	8.4	5.4
60 min.	---	13.4	9.9	3.8
90 min.	---	12.6	7.6	3.9
180 min.	---	13.0	8.6	5.6
Average Elution Assay (all times)	12.9	12.4	8.6	4.6
Ratio of Elution to Diffusion Assay	1.12	1.15	1.27	0.79**

*Impregnated product on disks were targeted to be 125% of this nominal potency.

**180 minute elution into water at 21 mos. and assay by the ultraviolet absorption method of CFR 141d.303 yielded 5.6 mcg. per disk (diffusion assay 5.1 mcg.), for an elution to diffusion ratio of 1.09 for lot 2015, and at 9 mos. an assay of 5.9 mcg. per disk, for an elution to diffusion ratio of 0.98 for lot 2457.

TABLE II

Cl Indamycin

Lot Numbers	2275	2275	2334	2355	2419
Manufactured	4/72	4/72	7/72	8/72	12/72
Mnfd. Nominal Conc.*, mcg.	2	2	2	2	2
Age at Assay	1 mo.	13 mos.	5 mos.	4 mos.	9 mos
Diffusion Assay	2.8	2.0	2.1	1.8	2.3
Elution Assay, 10 min.	2.0	2.0	1.5	1.8	1.8
20 min.	2.3	---	---	---	---
30 min.	2.3	2.0	1.4	1.7	2.0
60 min.	---	1.6	1.5	1.6	1.9
90 min.	---	1.9	1.5	1.8	1.8
180 min.	---	2.0	1.5	1.7	1.9
Average Elution Assay (all times)	2.2	1.9	1.5	1.7	1.9
Ratio of Elution to Diffusion Assay	0.78	0.95	0.72	0.94	0.82

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1E

Colistin

Lot Numbers	2252	2252	2463	2469
Manufactured	3/72	3/72	12/72	12/72
Mfd. Nominal Conc.*, mcg.	13	13	13	13
Age at Assay	3 wks.	10 mos.	2 mos.	1 mo.
Diffusion Assay	12.7	10.0	13.5	13.6
Elution Assay, 10 min.	17.6	11.1	11.8	10.0
20 min.	16.1	---	---	---
30 min.	16.3	10.0	10.7	10.2
60 min.	---	10.7	10.2	10.6
90 min.	---	9.9	10.1	10.1
180 min.	---	10.2	10.5	10.2
Average Elution Assay (all times)	16.7	10.4	10.6	10.2
Ratio of Elution to Diffusion Assay	1.30	1.04	0.78	0.75

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 11h - Part 1

Doxycycline

Lot Numbers	2384	2460	2466
Manufactured	10/72	12/72	12/72
Mnfd. Nominal Conc.*, mcg.	0.5	0.5	0.5
Age at Assay	4 mos.	1 mo.	1 mo.
Diffusion Assay	.61	.65	.71
Elution Assay, 10 min.	.29	.40	.40
20 min.	---	---	---
30 min.	.38	.53	.48
60 min.	.38	.60	.57
90 min.	.38	.54	.57
180 min.	.40	.59	.52
Average Elution Assay (60-180 min. values)	.39	.58	.55
Ratio of Elution to Diffusion Assay	0.64	0.89	0.77

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1h - Part 2

Doxycycline

Lot Numbers	2293	2293	2496	2500
Manufactured	5/72	5/72	1/73	1/73
Mfd. Nominal Conc.*, mcg.	2	2	2	2
Age at Assay	2 wks.	10 mos.	2 mos.	2 mos.
Diffusion Assay	2.3	2.2	2.7	2.9
Elution Assay, 10 min.	1.2	1.8	2.2	1.9
20 min.	1.2	---	---	---
30 min.	1.6	1.7	3.0	2.4
60 min.	---	1.7	2.4	2.7
90 min.	---	1.7	2.4	2.8
180 min.	---	1.8	2.2	3.0
Average Elution Assay (30-180 min. values)	1.6	1.7	2.5	2.8
Ratio of Elution to Diffusion Assay	0.70	0.78	0.92	0.96

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 11

Erythromycin

Lot Numbers	2258	2258	2443	2444
Manufactured	4/72	4/72	12/72	12/72
Mnfd. Nominal Conc.*, mcg.	2.5	2.5	2.5	2.5
Age at Assay	2 wks.	8 mos.	1 mo.	1 mo.
Diffusion Assay	3.3	3.3	3.0	3.0
Elution Assay, 10 min.	2.9	2.3	2.8	2.2
20 min.	2.8	---	---	---
30 min.	2.7	2.6	2.7	2.1
60 min.	---	2.6	2.4	2.0
90 min.	---	2.6	2.3	2.3
180 min.	---	2.1	1.9	2.0
Average Elution Assay (10-90 min. values)	2.8	2.5	2.5	2.2
Ratio of Elution to Diffusion Assay	0.85	0.78	0.83	0.74

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1j

Gentamicin

Lot Numbers	2255	2255	2464	2470	2525
Manufactured	4/72	4/72	12/72	12/72	2/73
Mufd. Nominal Conc.*, mcg.	9	9	9	9	9
Age at Assay	3 wks.	9 mos.	1 mo.	1 mo.	7 mos.
Diffusion Assay	8.5	8.5	11.4	10.9	11.9
Elution Assay, 10 min.	9.3	7.6	9.0	9.5	11.7
20 min.	9.1	---	---	---	---
30 min.	10.3	11.5	9.6	9.4	10.9
60 min.	---	8.2	10.0	11.1	11.5
90 min.	---	7.9	10.8	10.5	11.5
180 min.	---	7.4	9.7	8.1	11.1
Average Elution Assay (30-90 min. values)	9.6	8.9	10.1	10.3	11.3
Ratio of Elution to Diffusion Assay	1.17	1.05**	0.89	0.95	0.95

*Impregnated product on disks were targeted to be 125% of this nominal potency.

**A third assay on lot 2255 at 17 mos. of age gave a diffusion assay of 10.9 mcg., elution assay of 11.8; elution to diffusion ratio of 1.09.

TABLE 1k

Kanamycin

Lot Numbers	2261	2261	2486	2574
Manufactured	4/72	4/72	1/73	3/73
Mfrd. Nominal Conc.*, mcg.	18	18	18	18
Age at Assay	3 wks.	11 mos.	2 mos.	2 mos.
Diffusion Assay	18.6	19.8	22.1	21.4
Elution Assay, 10 min.	17.3	19.8	20.4	20.4
20 min.	20.9	---	---	---
30 min.	23.6	18.8	18.4	21.0
60 min.	---	17.3	18.3	18.6
90 min.	---	17.4	16.6	19.0
180 min.	---	18.2	14.4	18.3
Average Elution Assay (all times)	20.6	18.3	17.6	19.4
Ratio of Elution to Diffusion Assay	1.19	0.93	0.80	0.95

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE I I

Methicillin

Lot Numbers	2260	2260	2394	2471	2409
Manufactured	4/72	4/72	10/72	1/73	11/72
Std. Nominal Conc.*, mcg.	5	5	5	5	5
Age at Assay	2 wks.	10 mos.	3 mos.	2 wks.	10 mos.
Diffusion Assay	7.1	6.3	7.0	6.7	7.0
Elution Assay, 10 min.	5.9	4.5	4.9	4.7	6.0
20 min.	5.8	---	---	---	---
30 min.	5.7	4.5	5.5	4.2	5.5
60 min.	---	4.6	5.3	4.2	5.0
90 min.	---	4.5	4.7	4.1	5.8
180 min.	---	4.2	5.4	4.5	5.7
Average Elution Assay (all times)	5.8	4.5	5.2	4.3	5.6
Ratio of Elution to Diffusion Assay	0.82	0.71**	0.74	0.64	0.80

*Impregnated product on disks were targeted to be 125% of this nominal potency.

**A third assay on lot 2260 at 17 mos. yielded a diffusion assay of 6.3, and an elution assay of 5.3; elution to diffusion ratio of 0.83.

TABLE 1m

Nalidixic Acid

Lot Numbers	2288	2288	2425	2426
Manufactured	5/72	5/72	12/72	12/72
Mnfd. Nominal Conc.*, mcg.	15	15	15	15
Age at Assay	3 wks.	8 mos.	2 mos.	2 mos.
Diffusion Assay	19.4	19.4	21.7	22.4
Elution Assay(1) 10 min.	18.5	17.2	16.3	18.6
20 min.	18.6	---	---	---
30 min.	18.5	17.4	18.9	20.5
60 min.	---	18.9	19.4	20.0
180 min.	---	19.5	18.0	19.9
Average Elution Assay (all times)	18.5	18.6	18.8	20.1
Ratio of "Diffusion" to "Elution" Assay	0.98	0.95	0.87	0.90

*Impregnated production disks were targeted to be 125% of this nominal potency.

(1) By an ultraviolet spectrophotometric procedure.

TABLE In

Neomycin

Lot Numbers	2279	2674	2675	2676
Manufactured	5/72	6/73	6/73	4/73
Mnfd. Nominal Conc.*, mcg.	10	20	20	20
Age at Assay	2 wks.	3 mos.	3 mos.	3 mos.
Diffusion Assay	9.5	29	26	29
Elution Assay, 10 min.	6.4	13.2	15.7	16.0
20 min.	7.3	---	---	---
30 min.	8.2	15.4	15.9	15.9
60 min.	---	17.7	15.9	13.5
90 min.	---	14.2	17.2	12.9
180 min.	---	20.6	16.5	14.0
Average Elution Assay (all times)	7.3	16.2	16.2	15.9
Ratio of Elution to Diffusion Assay	0.77	0.56	0.63	0.55

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1o

Nitrofurantoin

Lot Numbers	2287	2287	2534	2659	2662	2663
Manufactured	5/72	5/72	2/73	6/73	6/73	6/73
Mat'l. Nominal Conc.*, mcg.	25	25	15	15	15	15
Age at Assay	1 mo.	8 mos.	7 mos.	3 wks.	3 wks.	3 wks.
Diffusion Assay(1)	30.3	28.9	21.0	21.1	19.0	22.9
Elution Assay(2), 10 min.	31.8	27.1	---	---	---	---
20 min.	33.0	---	---	---	---	---
30 min.	36.9	28.1	19.2	18.0	18.7	17.7
60 min.	---	29.1	---	18.6	---	---
180 min.	---	28.5	---	---	---	---
Average Elution Assay (all times)	33.9	28.2	19.2	18.3	18.7	17.7
Ratio of Elution to Diffusion Assay	1.12	0.97	0.91	0.87	0.98	0.78

*Impregnated product on disks were targeted to be 125% of this nominal potency.

(1)The "diffusion" assay was a microbiological procedure described elsewhere.

(2)By an ultraviolet spectrophotometric procedure.

TABLE 1p

Novobiocin

Lot Numbers	2269	2269	2447	2448
Manufactured	4/72	4/72	12/72	12/72
Mufd. Nominal Conc.*, mcg.	2.5	2.5	2.5	2.5
Age at Assay	2 wks.	11 mos.	3 mos.	3 mos.
Diffusion Assay	1.8	1.4	3.0	3.2
Elution Assay, 10 min.	0.6	0.4	1.8	1.6
20 min.	0.7	---	---	---
30 min.	0.8	0.5	2.1	1.9
60 min.	---	0.6	2.2	1.6
90 min.	---	0.5	2.0	1.9
180 min.	---	0.6	2.1	1.9
Average Elution Assay (30-180 min. values)	0.8	0.6	2.1	1.8
Ratio of Elution to Diffusion Assay	0.45	0.43	0.70	0.56

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1q

Oleandomycin

Lot Numbers	2256	2256	2441	2442
Manufactured	3/72	3/72	12/72	12/72
Mfd. Nominal Conc.*, mcg.	7.5	7.5	7.5	7.5
Age at Assay	4 wks.	12 mos.	4 mos.	4 mos.
Diffusion Assay	9.2	8.2	9.2	8.9
Elution Assay, 10 min.	10.9	9.9	11.5	12.0
20 min.	12.3	---	---	---
30 min.	15.2	9.3	12.8	12.2
60 min.	---	9.8	12.1	12.3
90 min.	---	10.3	12.5	12.7
180 min.	---	9.7	12.8	12.5
Average Elution Assay (all times)	12.8	9.8	12.3	12.3
Ratio of Elution to Diffusion Assay	1.38	1.20	1.33	1.38

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1r

Penicillin

Lot Numbers	2267	2385	2461	2467	2368
Manufactured	4/72	9/72	12/72	12/72	8/72
Mfd. Nominal Conc.*, U.	0.4	0.2	0.2	0.2	0.2
Age at Assay	9 mos.	3 mos.	1 mo.	1 mo.	11 mo.
Diffusion Assay	0.52	0.24	0.28	0.28	0.26
Elution Assay, 10 min.	0.39	0.18	0.22	0.25	0.18
20 min.	---	---	---	---	---
30 min.	0.40	0.21	0.24	0.25	0.18
60 min.	---	0.18	0.23	0.23	0.17
90 min.	---	0.18	0.23	0.20	0.17
180 min.	---	0.16	0.22	0.21	0.18
Average Elution Assay (all times)	0.40	0.18	0.23	0.23	0.18
Ratio of Elution to Diffusion Assay	0.77**	0.75	0.82	0.82	0.69

*Impregnated product on disks were targeted to be 125% of this nominal potency.

**A second assay on lot 2267 at 17 mos. showed a diffusion assay of 0.41, an elution assay of 0.28; elution to diffusion ratio of 0.69.

TABLE 1s

Polymyxin B

Lot Numbers	2285	2285	2482	2483
Manufactured	5/72	5/72	1/73	1/73
Mnfd. Nominal Conc.*, U.	12.5	12.5	12.5	12.5
Age at Assay	2 mos.	10 mos.	2 mos.	2 mos.
Diffusion Assay	11.6	13.9	16.0	15.6
Elution Assay[1] 10 min.	---	3.1	3.5	5.2
20 min.	---	---	---	
30 min.	---	5.8	8.7	6.8
60 min.	6.8	7.2	7.7	8.4
90 min.	---	7.6	9.5	9.9
120 min.	7.6			
180 min.	8.0	8.9	10.0	9.5
Average Elution Assay (180 min. value only)	8.0	8.9	10.0	9.5
Ratio of Elution to Diffusion Assay	0.69	0.64	0.63	0.61

*Impregnated product on disks were targeted to be 125% of this nominal potency.

(1) The 'elution assay' was conducted by a modified diffusion assay on the disk after elution for the time designated. See text.

TABLE It

Streptomycin

Lot Numbers	2280	2280	2495	2498	2679	2680
Manufactured	5/72	5/72	1/73	1/73	6/73	6/73
Mold. Nominal Conc.*, mcg.	10	10	20	20	20	20
Age at Assay	4 wks.	11 mos.	3 mos.	3 mos.	3 mos.	3 mos.
Diffusion Assay	11.4	12.6	21.2	22.5	21.1	27.1
Elution Assay, 10 min.	8.3	9.5	17.3	16.8	14.8	19.5
20 min.	9.0	---	---	---	---	---
30 min.	10.3	9.4	16.4	16.8	15.4	20.5
60 min.	---	9.5	18.0	15.6	19.7	17.4
90 min.	---	9.3	17.4	18.7	15.2	19.9
180 min.	---	10.4	17.4	16.7	13.6	18.3
Average Elution Assay (all times)	9.2	9.6	17.3	16.9	15.7	19.2
Ratio of Elution to Diffusion Assay	0.81	0.76	0.82	0.75	0.74	0.71

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1u - Part 1

Tetracycline

Lot Numbers	2373	2462	2468
Manufactured	9/72	12/72	12/72
Mufd. Nominal Conc.*, mcg.	0.5	0.5	0.5
Age at Assay	5 mos.	1 mo.	1 mo.
Diffusion Assay	0.60	0.67	0.60
Elution Assay, 10 min.	0.37	0.42	0.46
20 min.	---	---	---
30 min.	0.54	0.54	0.44
60 min.	0.45	0.53	0.47
90 min.	0.48	0.55	0.49
180 min.	0.45	0.55	0.50
Average Elution Assay (30-180 min. values)	0.48	0.54	0.48
Ratio of Elution to Diffusion Assay	0.80	0.81	0.80

*Impregnated production disks were targeted to be 125% of this nominal potency.

TABLE 1a - Part 2

Tetracycline

Lot Numbers	2278	2278	2497	2499
Manufactured	4/72	4/72	1/73	1/73
Mfd. Nominal Conc.*, mcg.	1.2	1.2	1.2	1.2
Age at Assay	2 wks.	9 mos.	1 mo.	1 mo.
Diffusion Assay	1.38	1.17	1.22	1.20
Elution Assay, 10 min.	1.08	.75	1.00	0.85
20 min.	1.05	---	---	---
30 min.	1.08	.98	1.01	1.11
60 min.	---	.89	0.82	1.01
90 min.	---	.81	1.08	1.08
180 min.	---	.92	0.86	1.07
Average Elution Assay (30-180 min. values)	1.08	0.90	0.89	1.07
Ratio of Elution to Diffusion Assay	0.78	0.77	0.73	0.89

Note: Additional elution assays, on a 5 mcg. disk, are presented in Table 6.

*Impregnated product on disks were targeted to be 125% of this nominal potency.

TABLE Iv

Vancomycin

Lot Numbers	2276	2276	2427	2428
Manufactured	4/72	4/72	12/72	12/72
Mfrd. Nominal Conc.*, mcg.	10	10	10	10
Age at Assay	4 wks.	8 mos.	1 mo.	1 mo.
Diffusion Assay	9.8	9.9	13.6	12.3
Elution Assay, 10 min.	13.7	9.3	15.6	14.5
20 min.	13.5	---	---	---
30 min.	12.7	11.0	15.8	14.7
60 min.	---	10.1	14.5	13.3
90 min.	---	12.6	12.8	12.7
180 min.	---	10.2	15.0	17.6
Average Elution Assay (all times)	13.3	10.6	14.7	14.2
Ratio of Elution to Diffusion Assay	1.35	1.07	1.06	1.15

*Impregnated production disks were targeted to be 125% of this nominal potency.

Elution/Diffusion Assay Ratios for Various Disks

Disk	Manufacturer's Nominal Potency, mcg. (U.)	Lot Number	Ratio Elution/Diffusion Avg. ± SD, SD x t_{90}
Ampicillin	0.25	2354	.89
		2459	.79
		2465	.88
		Avg.	.85 ± .045, .13
Ampicillin	5.0	2330	.76
		2650	.66
		2736	.75
		Avg.	.72 ± .045, .13
Ampicillin	5	2266	.92
			.70
		2421	.83
		2422	.76
		Avg.	.80 ± .08, .19
Bacitracin	12 U.	2239	.86
			1.17
		2445	1.31
		2446	1.25
		Avg.	1.15 ± .17, .40
Bacitracin	18 U.	2703	1.27
		2704	1.30
		2705	1.22
		Avg.	1.26 ± .03, .09
Carbenicillin	120	2291	.61
			.75
		2531	.68
		2577	.67
		Avg.	.68 ± .05, .12
Cephalothin	15	2271	1.06
			.98
			.83
		2423	.95
		2424	.99
		2532	.77
		Avg.	.93 ± .11, .21

Elution/Diffusion Assay Ratios for Various Disks

Disk	Manufactured Nominal Potency, mcg.(U.)	Lot Number	Ratio Elution/Diffusion* Avg. ± SD, SD x t_{90}
Chloramphenicol	5	2253	1.12
			1.15
		2015	1.27
		2457	0.79
		Avg.	1.08 ± .18, .42
Clindamycin	2	2275	.78
			.95
		2334	.72
		2335	.94
		2419	.82
		Avg.	.84 ± .09, .19
Colistin	13	2252	1.30
			1.04
		2463	.78
		2469	.75
		Avg.	.97 ± .22, .52
Doxycycline a	0.5	2384	.64
		2460	.89
		2466	.77
		Avg.	.77 ± .10, .30
Doxycycline	2.0	2293	.70
			.78
		2496	.92
		2500	.96
		Avg.	.84 ± .11, .25
Erythromycin	2.5	2258	.85
			.78
		2443	.83
		2444	.74
		Avg.	.80 ± .04, .10
Gentamicin	9	2255	1.17
			1.05
			1.09
		2464	.89
		2470	.95
		2525	.95
		Avg.	0.95 ± .10, .19

SECTION 8 PAGE

Elution/Diffusion Assay Ratios for Various Disks

Disk	Manufactured Nominal Potency, mcg.(U.)	Lot Number	Ratio Elution/Diffusion* Avg. ± SD, SD × t_{90}
Kanamycin	1S	2261	1.19 .93
		2486	.80
		2574	.95
		Avg.	.97 ± .14, .33
Methicillin	5	2260	.82 .71 .83
		2394	.74
		2409	.80
		2471	.64
		Avg.	.76 ± .07, .14
Nalidixic Acid	15	2288	.98 .95
		2425	.87
		2426	.90
		Avg.	.93 ± .04, .10
Neomycin	10,20	2279	.86
		2674	.56
		2675	.63
		2676	.55
		Avg.	.65 ± .13, .29
Nitrofurantoin	25,15	2287	1.12 .97
		2534	.91
		2659	.87
		2662	.99
		2663	.93
		Avg.	.94 ± .12, .31
Novobiocin	2.5	2269	.45 .43
		2447	.70
		2448	.56
		Avg.	.54 ± .11, .25

Elution/Diffusion Assay Ratios for Various Disks

Disk	Manufactured Nominal Potency, mcg. (U.)	Lot Number	Ratio Elution/Diffusion* Avg. ± SD, SD x t_{90}
Oleandomycin	7.5	2556	1.38
			1.20
		2441	1.33
		2442	1.38
		Avg.	1.32 ± .07, .17
Penicillin G	0.2 U.	2385	.75
		2461	.82
		2467	.82
		2368	.62
		Avg.	.75 ± .08, .19
Polymyxin B	12.5 U.	2285	.69
			.64
		2482	.63
		2483	.61
		Avg.	.64 ± .03, .07
Streptomycin	10,20	2280	.81
			.76
		2495	.82
		2498	.75
		2679	.74
		2680	.71
		Avg.	.77 ± .04, .08
Tetracycline	0.5	2373	.80
		2462	.81
		2468	.80
		Avg.	.80 ± .004, .01
Tetracycline	1.2	2278	.78
			.77
		2497	.73
		2499	.89
		Avg.	.79 ± .06, .14
Vancomycin	10	2276	1.35
			1.07
		2427	1.06
		2428	1.15
		Avg.	1.16 ± .12, .27

Elution/Diffusion Assay Ratios for Various Disks

*The 90% confidence limits calculated for the ratios are based on the appropriate Student's t factor, corresponding to the number of assays listed. The term "90%" must be considered nominal, however, since the precise probability distribution characteristics of the ratio have not been established.

Precision of Antibiotic Assays

Antibiotic	Manufactured Nominal Potency, mcg. (f.)	Lot Number	Coefficient of Variation, % Diffusion (n=12)[1]	Elution (n=8)[2,3]
Ampicillin	7.5	2266	14	24
		2421	10	32
		2422	13	26
		Avg.	12	27
Ampicillin	5.0	2530	14	19
		2660	8	22
		2736	15	16
		Avg.	12	19
Ampicillin	0.25	2354	17	13
		2459	29	9
		2465	37	11
		Avg.	28	11
Bacitractin	10 U.	2239	11	13
		2445	9	11
		2446	9	11
		Avg.	10	12
Bacitracin	18 U.	2703	19	10
		2704	19	13
		2705	19	14
		Avg.	19	12
Carbenicillin	120	2291	15	11
		2531	16	10
		2577	16	6
		Avg.	16	9
Cephalothin	15	2271	9	26
		2423	14	24
		2424	21	26
		2532	7	22
		Avg.	13	25
Chloramphenicol	5	2015	10	17
		2253	8	23
		2340	8	26
		2457	-	37
		Avg.	9	26

Precision of Antibiotic Assays

Antibiotic	Manufactured Nominal Potency, mcg. (U.)	Lot Number	Coefficient of Variation, % Diffusion (n=12)[1]	Elution (n=8)[2,3]
Clindamycin	2	2275	15	26
		2334	15	16
		2355	22	24
		2419	14	13
		Avg.	16	20
Colistin	13	22[illegible]2	16	16
		2463	14	14
		2469	12	12
		Avg.	14	14
Doxycycline	0.5	2384	11	18
		2460	19	28
		2466	15	31
		Avg.	15	26
Doxycycline	2	2293	29	17
		2496	11	32
		2500	9	35
		Avg.	16	28
Erythromycin	2.5	2258	27	29
		2443	18	37
		2444	18	20
		Avg.	21	29
Gentamicin	9	2255	19	34
		2464	14	25
		2470	11	32
		2525	13	18
		Avg.	14	27
Kanamycin	18	2261	13	21
		2486	8	20
		2487	9	17
		Avg.	10	19
Methicillin	5	2260	4	11
		2394	6	18
		2409	19	17
		2471	6	16
		Avg.	9	15

Precision of Antibiotic Assays

Antibiotic	Manufactured Nominal Potency, mcg. (U.)	Lot Number	Coefficient of Variation, % Diffusion (n=12)[1]	 Elution (n=8)[2,3]
Nalidixic Acid	15[4]	2288	12	9
		2425	11	6
		2426	13	8
		Avg.	12	8
Neomycin	20	2674	10	25
		2675	14	10
		2676	7	18
		Avg.	8	18
Nitrofurantoin	15[4]	2659	9	8
		2662	7	6
		2663	8	6
		Avg.	8	7
Novobiocin	2.5	2269	21	27
		2447	19	16
		2448	32	14
		Avg.	24	19
Oleandomycin	7.5	2256	18	13
		2441	30	14
		2442	23	14
		Avg.	24	14
Penicillin	0.2 U.	2368	7	19
		2385	23	36
		2461	7	35
		2467	10	38
		Avg.	12	32
Polymyxin B[5]	12.5 U.	2285	11	16
		2482	8	42
		2483	20	21
		Avg.	13	28
Streptomycin	20	2495	11	11
		2498	32	12
		2679	21	21
		2680	19	23
		Avg.	21	19

TABLE 3

Precision of Antibiotic Assays

Antibiotic	Manufactured Nominal Potency, mcg. (U.)	Lot Number	Coefficient of Variation, % Diffusion (n=12.)[1]	Elution (n=8)[2,3]
Tetracycline	0.5	2373	23	21
		2462	28	22
		2468	11	17
		Avg.	21	20
Tetracycline	1.2	2278	10	20
		2497	13	15
		2499	18	18
		Avg.	14	18
Vancomycin	10	2276	12	25
		2427	9	26
		2428	11	23
		Avg.	11	25
		Overall Avg.	15	19

FOOTNOTES

TABLE 3

Precision of Antibiotic Assays

[1]The precision of diffusion assays reported here is in general accord with our long-term experience in antibiotic susceptibility disk assays by CFR methods.

[2]The precision of elution assays is usually lower than that of diffusion assays. We can identify the following sources of error in elution assays in addition to those which occur in diffusion assays: a) variations in total volume of eluant used from run to run, b) variations in volume of eluant from disk to disk, due to compartment variation, c) additional errors inherent in dilution of eluate, d) additional errors in measuring eluant to cups which were filled to brim, not volumetrically.

[3]For elution assays, the coefficients of variation given are calculated from the total of all assays at all times; thus, for those antimicrobial agents which show incomplete elution at early times, or degradation at later times, the coefficient of variation recorded is higher than for individual time measurements. This is especially true for Cephalothin, Erythromycin, Gentamicin, Novobiocin and Penicillin G.

[4]Ultraviolet spectrophotometric assay methods, inherently more accurate than bioassays, were used for Nalidixic Acid and Nitrofurantoin elution studies, hence the low coefficients of variation for these.

[5]For Polymyxin B all assays were conducted by diffusion methods. The coefficient of variation quoted for the "elution" assay is for the 180 minute period only.

STATISTICAL ANALYSIS OF ASSAY VARIABILITY

Summary

The number of dilutions, plates, and production discs used in the Autobac assay clearly has an effect on assay precision. Accordingly an experiment was designed to estimate the contribution of each of these sources to the assay variability. For each antibiotic, 10 dilutions, 3 plates per dilution, and 6 discs per plate were used. An analysis-of-variance model was used to estimate the magnitude of the three components of variation.

From these variation estimates it is possible to compute an estimated measure of precision in the ultimate (production) assay, as a function of the number of dilutions, plates, and discs ultimately to be used. Tables assuming various values for these parameters are given. The measure of precision is an upper bound on the error such that the error would fall short of this bound 95% of the time. Here "error" means the difference between the assayed potency of the sample and the assayed potency one would get if one assayed the entire lot. It is expressed as a percent of the mean potency and refers to error in either direction (i.e. it is a plus-or-minus figure).

Analysis of Variance

Mathematical model: $y_{ijk} = \mu + \alpha_i + \beta_{(i)j} + \gamma_{(ij)k}$

where y_{ijk} = assayed potency of disc ijk

μ = overall mean potency

α_i = effect of i^{th} dilution*

$\beta_{(i)j}$ = effect of ij^{th} plate (stand. discs)

$\gamma_{(ij)k}$ = effect of ijk^{th} (production) disc

The "analysis-of-variance table" looks like this (for Ampicillin):

Source of Variation	d.f.	Mean Square	Expected Mean Square
Dilutions	9	3.7798	$\sigma^2_\gamma + 6\sigma^2_\beta + 18\sigma^2_\alpha$
Plates (within Dilutions)	20	1.4555	$\sigma^2_\gamma + 6\sigma^2_\beta$
Discs (within Plates)	150	0.6351	σ^2_γ
Total	179		

The α_i, $\beta_{(i)j}$ and $\gamma_{(ij)k}$ in the mathematical model above are assumed to be random, each with mean value 0 (so that μ is the mean for y) and with variances σ^2_α, σ^2_β and σ^2_γ respectively.*

We use the observed Mean Squares as estimates of the expected Mean Squares and obtain (where σ^2 now indicates an estimated variance):

$$\sigma^2_\gamma + 6\sigma^2_\beta + 18\sigma^2_\alpha = 3.7798$$

$$\sigma^2_\gamma + 6\sigma^2_\beta = 1.4555$$

$$\sigma^2_\gamma = 0.6351$$

STATISTICAL ANALYSIS OF ASSAY VARIABILITY

Analysis-of-Variance (continued)

These equations solve easily to give $\sigma_\alpha^2 = 0.1291$

$\sigma_\beta^2 = 0.1367$

$\sigma_\gamma^2 = 0.6351$

These three values are the (estimated) variance of the effect of a dilution, a plate, and a disc, respectively.

Variability in Production

In production you will use a dilutions, b plates per dilution, and c (production) discs per plate, and will average the resulting abc potencies to obtain a sample mean

$$\bar{y} = \frac{1}{abc} \sum_i \sum_j \sum_k y_{ijk}$$

$$= \frac{1}{abc} \sum_i \sum_j \sum_k (\mu + \alpha_i + \beta_{(i)j} + \gamma_{(ij)k})$$

$$= \mu + \frac{1}{a} \sum_i \alpha_i + \frac{1}{ab} \sum_i \sum_j \beta_{(i)j} + \frac{1}{abc} \sum_i \sum_j \sum_k \gamma_{(ij)k}$$

whose variance is

$$V(\bar{y}) = \frac{1}{a} \sigma_\alpha^2 + \frac{1}{ab} \sigma_\beta^2 + \frac{1}{abc} \sigma_\gamma^2$$

The expectation of $\bar{y}$ is μ. Note that μ is the mean assayed potency of the entire population of discs, while $\bar{y}$ is the mean of a sample from this population.

Using the estimates of σ_α^2, σ_β^2 and σ_γ^2 obtained above, one uses the immediately preceding formula to estimate the variance of $\bar{y}$. The square root of this variance is then the standard deviation of $\bar{y}$, $\sigma_{\bar{y}}$.

The error $\bar{y}-\mu$ satisfies

$$|\bar{y}-\mu| \leq 1.96\sigma_{\bar{y}}$$

with probability 0.95. Thus $1.96\sigma_{\bar{y}}$ is a 95% confidence upper bound for the magnitude of the error. This bound can be converted to a percent figure by dividing by the mean value obtained for y in the study. This percent figure is tabled below.

The analysis-of-variance tables and the tables of percent error bounds are given below. In the case of doxycycline the F-test for dilutions is not quite statistically significant, but in the interest of caution its estimated contribution to variability has been included anyway.

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*The mathematical model used for the analysis-of-variance is that of a one-way random-effects nested design, see e.g. G. W. Snedecor and W. G. Cochran, "Statistical Methods," 6th ed., Iowa State University Press, Ames (1967) 285-88. We have of course assumed throughout that the random effects are normally distributed.

ANALYSIS OF VARIANCE TABLE — AMPICILLIN

	DF	SUM SQ	MEAN SQ	F
DILUTIONS	9	34.0179	3.7798	2.60*
PLATES W. DILUT	20	29.1103	1.4555	2.29**
DISCS W. PLATES	150	95.2607	0.6351	
TOTAL	179	158.3888		

* $p < 0.05$

** $p < 0.01$

ESTIMATES OF COMPONENTS OF VARIANCE

$$\sigma^2_{\gamma}(\text{INC}) = \frac{3.7798 - 1.4555}{18} = 0.1291$$

$$\sigma^2_{\beta}(\text{PLA}) = \frac{1.4555 - 0.6351}{6} = 0.1367$$

$$\sigma^2_{\delta}(\text{DIS}) = 0.6351$$

MEAN SQUARE = 1.5541

N 50580 -8

a	b	c	90% confidence error bound
1	5	6	18.1 %
2	5	6	12.8
3	5	6	10.5
4	5	6	9.1
5	5	6	8.1
6	5	6	7.4
7	5	6	6.9
8	5	6	6.4
9	5	6	6.0
10	5	6	5.7
1	10	6	16.9
2	10	6	11.9
3	10	6	9.7
4	10	6	8.4
5	10	6	7.5
6	10	6	6.9
7	10	6	6.4
8	10	6	6.0
9	10	6	5.6
10	10	6	5.3

ANALYSIS OF VARIANCE TABLE — Dobyns case

	DF	SUM SQ	MEAN SQ	F
[illegible]	9	3.7072	0.4119	2.05*
[illegible]	20	4.0268	0.2013	9.62***
[illegible]	150	3.1394	0.0209	
[illegible]	179	10.8734		

* p < 0.10

*** p < .001

ESTIMATES OF [illegible] OF VARIANCE

σ^2_u (BIL) = $\frac{0.4119 - 0.2013}{15}$ = 0.01170

$\sigma^2_{u(BIL)}$ (PCA) = $\frac{0.2013 - 0.0209}{6}$ = 0.03007

σ^2_e (DIS) = 0.02093

AND [illegible] = 1.517

1	5	6	16.7
2	5	6	11.8
3	5	6	9.6
4	5	6	8.3
5	5	6	7.4
6	5	6	6.8
7	5	6	6.3
8	5	6	5.9
9	5	6	5.6
10	5	6	5.3
1	10	6	15.1
2	10	6	10.6
3	10	6	8.7
4	10	6	7.5
5	10	6	6.7
6	10	6	6.1
7	10	6	5.7
8	10	6	5.3
9	10	6	5.0
10	10	6	4.8

Antimicrobial Agent	Nominal Potency	Optimal Potency Range, % of Nominal	Number of Curves	Number of Plates on Curve	Number of Disks on Plate
ampicillin	3.6 mcg.	80-125	3	5	6
ampicillin	0.22 mcg.	80-130	3	5	6
bacitracin	18 U.	80-150	1	3	2
carbenicillin	120 mcg.	80-150	1	3	2
cephalothin	15 mcg.	68-150	1	3	2
chloramphenicol	4 mcg.	80-150	1	3	2
clindamycin	2 mcg.	68-150	1	3	2
colistin	13 mcg.	68-150	1	3	2
doxycycline	1.6 mcg.	80-130	3	5	6
doxycycline	0.5 mcg.	68-130	1	3	2
erythromycin	2.5 mcg.	68-150	1	3	2
fusidic	9 mcg.	68-130	1	3	2
gentamycin	22 mcg.	80-150	1	3	2
kanamycin	2.4 mcg.	68-150	1	3	2
methicillin	5 mcg.	68-130	1	3	2
nalidixic Acid*	15 mcg.	68-130	1	3	2
neomycin	23 mcg.	68-150	1	3	2
nitrofurantoin*	15 mcg.	80-125	3	5	6
novobiocin	2.5 mcg.	68-130	1	3	2
oxytetracycline	6 mcg.	68-150	1	3	2
penicillin G	0.2 U.	68-130	1	3	2
polymyxin B	12.5 U.	68-150	1	3	2
streptomycin	20 mcg.	80-150	1	3	2

*V. assay is preferred.

COPY FROM FILE

A Microbiological Assay Procedure for the Assay of Certain Antibiotic Fermentation Broths for the Autobac 1

Antimicrobial Agent	Nominal Potency	Optimal Potency Range, % of Nominal	Number of Curves	Number of Plates per Curve	Number of Dilutions per Plate
Tetracycline	1.2 mcg.	80-130	3	5	6
Tetracycline	0.5 mcg.	68-150	1	3	2
Vancomycin	10 mcg.	68-180	1	3	2

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TABLE 6

Methodology of Cup-Plate (Elution) Assay of Autobac 1
Elution Disks, and Comparison to CFR Method

Antibiotic	CFR Reference	Base Layer Volume CFR	Base Layer Volume Pfizer	CFR Organism	Pfizer Organism	Organism Volume CFR	Organism Volume Pfizer	Standard Curve Range, mcg./(U.) CFR	Standard Curve Range, mcg./(U.) Pfizer	Diluent for Standard CFR	Diluent for Standard Pfizer[d]	Validity from CFR Method
Ampicillin	141a.111	21ml	21ml	S. lutea	S. lutea	0.5ml%	0.5ml%	0.064-0.156	0.06-0.15	buffer	buffer [illegible]	
Bacitracin	141e.401	21ml	21ml	M. flavus	M. flavus	0.3ml%	0.3ml%	0.25-0.6	0.25-0.6	buffer	buffer	
Carbenicillin	149y.1	21ml	21ml	P.aeruginosa	P.aeruginosa	0.5ml%	0.05ml%	12.8-31.2	6-15	buffer	[illegible]	[illegible]
Cephalothin	145w.1	21ml	21ml	S. aureus	S. aureus	0.1ml%	0.75ml%[a]	0.64-1.56	0.6-1.5	buffer	buffer	[illegible]
Chloramphenicol	141d.301	21ml	10ml	S. lutea	S. lutea	1.5ml%	1.2ml%	3-30	3-30	buffer	saline-broth	[illegible]
Clindamycin	149u.1	21ml	21ml	S. lutea	S. lutea	1.5ml%	1ml%	0.64-1.56	0.6-1.5	buffer	saline-broth	[illegible]
Colistin	148c.1	21ml	21ml	B. bronch.	B. bronch.	0.1ml%	0.1ml%	0.64-1.56	0.6-1.5	buffer	buffer	
Doxycycline	148z.1	21ml	10ml	B. cereus	B. cereus	test plate	test plate	0.64-1.56	0.15-0.4	buffer	buffer [illegible] saline-broth .5mcg/ml	[illegible]
Erythromycin	148e.1	21ml	21ml	S. lutea	S. lutea	1.5ml%	1ml%	0.64-1.56	0.6-1.5	buffer	saline-broth	[illegible]
Gentamicin	148q.1	21ml	21ml	S. epiderm	S. epiderm	1.5ml%	0.1ml%[b]	0.64-1.56	0.6-1.5	buffer	buffer	[illegible]
Kanamycin	148h.1	21ml	21ml	S. lutea	S. aureus	0.4ml%	0.5ml%	3.2-7.8	3-8	buffer	saline-broth	[illegible]
[illegible]	148x.1	21ml	21ml	S. lutea	S. lutea	1.5ml%	(c)	1.28-2.50	(c)	buffer	[illegible]	[illegible]
Methicillin	148a.103	21ml	21ml	S. aureus	S. lutea	0.3-1.0ml%	0.2ml%	3.12-15.6	3.0-1.5	buffer	saline-broth	[illegible]
Neomycin	148f.1	21ml	21ml	S. epiderm.	S. epiderm	0.4ml%	0.2ml%	0.64-1.56	0.6-1.5	buffer	buffer	
Novobiocin	148j.1a	21ml	21ml	S. epiderm	S. epiderm	4ml%	0.75ml%	0.32-0.78	0.3-0.8	buffer	saline-broth	[illegible]
Oleandomycin	148m.1	21ml	21ml	S. epiderm	S. epiderm	1ml%	0.5ml%	3.2-7.8	3-8	buffer	saline-broth	[illegible]
Penicillin	141a.1	21ml	21ml	S. aureus	S. lutea	1ml%	0.1ml%	0.04-4	0.06-0.15	buffer	saline-broth	[illegible]
Polymyxin B	148p.1	21ml	--	B. broneh.	--	0.1ml%	--	6.4-15.6	--	buffer	Modified Assay Used	
Streptomycin	141b.101	21ml	21ml	B. subtilis	B. subtilis	test plate	test plate	0.64-1.56	0.6-1.5	buffer	buffer	
Tetracycline	141c.218	21ml	10ml	B. cereus	B. cereus	test plate	test plate	0.64-1.56	0.15-0.4	buffer	buffer (1.2mcg./) saline-broth .5mcg./ml	[illegible]
Vancomycin	148s.1	21ml	21ml	B. cereus	S. epiderm		0.8ml%	6.4-15.6	2.5-10.0	buffer	saline-broth	[illegible]

(a) 60% T, not 25% designated by CFR.

(b) 0.1 ml of a 1:10 dilution of a 25% T suspension, not 1:4 dilution as designated by CFR.

(c) Elution assay not conducted as of this tabulation.

(d) Diluent the same for all disk potencies of a given antibiotic unless otherwise specified in this column.

Standard Curve for Diffusion Assays Performed on Elution Disks

Disk	Potency mcg(U)	Code		Standard Curve Points used, mcg(U)					Differs from CFR
Ampicillin (1)	0.22	AM	ep	.05	.10	.25	.5	1.0	Yes
Ampicillin (1)	1.6	AM	en	1.3	2.4	4.4	8.1	15.0	No
Bacitracin	18	B	ep	3.3	6.3	12.2	23.4	45	No
Carbenicillin	120	CB	en	33	63	122	234	450	Yes
Cephalothin	15	CL	ea	15	21.2	30.0	42.4	60.0	No
Chloramphenicol	4	C	en	3.3	6.3	12.2	23.4	45.0	No
Clindamycin	2	CM	ep	0.33	0.63	1.22	2.34	4.50	Yes
Colistin	13	CS	en	1.3	2.4	4.4	8.1	15.0	No
Doxycycline	0.5	DX	ep	0.33	0.63	1.22	2.34	4.50	Yes
Doxycycline	1.6	DX	en	"	"	"	"	"	"
Erythromycin	2.5	E	ep	1.30	2.70	5.40	11.0	22.5	No
Gentamicin	9	GM	ea	1.3	2.4	4.4	8.1	15.0	No
Kanamycin	22	K	en	3.3	6.3	12.2	23.4	45.0	No
Methicillin	5	SC	ep	1.3	2.4	4.4	8.1	15.0	No
Nalidixic Acid	15	NA	en	3	6	12	24	48	N.R.(2)
Neomycin	24	N	en	3.3	6.3	12.2	23.4	45.0	No
Nitrofurantoin	15	FD	en	3	6	12	24	48	N.R.(2)
Novobiocin	2.5	NV	ep	1.00	1.41	2.00	2.82	4.00	Yes
Oleandomycin	6	OL	ep	1.3	2.7	5.4	11.0	22.5	No
Penicillin G(1)	0.2U	P	ep	.05	.10	.20	.50	1.0	Yes
Polymyxin B	12.5U	PB	en	10	12.5	15	20	25 (3)	Yes
Streptomycin	20	ST	en	3.3	6.3	12.2	23.4	45.0	Yes
Tetracycline	0.5	TE	ep	0.33	0.63	1.22	2.34	4.50	Yes
	1.2	TE	en	"	"	"	"	"	"
Vancomycin	10	VA	ep	3.3	6.3	12.2	23.4	45.0	No

(1) See separate report for details of assay

(2) N.R. - No CFR method exists. Our assay procedure for Nitrofurantoin and Nalidixic Acid resemble the CFR 147.1 method. Details are presented under "Ancillary Data."

(3) Tentative values, to be confirmed.

TABLE 6

Effect of Disk Age on Elution to Diffusion Assay Ratio
(Based on 10 minute Elution Assay Value only)

Antibiotic	Manufactured Nominal Potency, mcg.	Lot No.	Approximate Age at Assay and Elution/Diffusion - Ratio		
Ampicillin	7.5*	2266	2 wks. 7.4/8.4=.88	8 mos. 5.9/8.7=.68	
	10†	2073			14 mos. 8.3/10.4=.80
Bacitracin	10U†	2239	2 wks. 10.4/12.4=.84		8 mos. 13.4/12.4=1.
		2446	2 wks. 16.2/13.2=1.23		
	18U	2703		2 mos. 25.6/20.3=1.27	
Carbenicillin	120	2291	2 wks. 87/144=.61		12 mos. 88/117=.75
	50†	2198		4 mos. 46/72=.64	
Cephalothin	15	2271	2 wks. 17.6/17.7=1.0	9 mos. 18.4/18.6=1.0	17 mos. 15.4/17.2=0.9
	30†	2178	6 mos. 35/37=.95		
Chloramphenicol	7.5*	2253	8 wks. 11.4/11.5=.99	8 mos. 10.7/10.8=.99	
	5†	2015			23 mos. 8.7/6.8=1.27
Clindamycin	2^	2275	1 mo. 2.0/2.8=.72		13 mos. 2.0/2.0=1.0
		2419		9 mos. 1.8/2.3=.78	
Colistin	13	2252	3 wks. 17.6/12.7=1.38	10 mos. 11.1/10.0=1.11	
	10†	2064			14 mos. 20.5/14.4=1.4

Effect of Disk Age on Elution to Diffusion Assay Ratio
(Based on 10 minute Elution Assay Value only)

Antibiotic	Manufactured Nominal Potency, mcg.	Lot No.	Approximate Age at Assay and Elution/Diffusion = Ratio		
Doxycycline	2*	2293	2 wks. 1.2/2.3=.52	10 mos. 1.8/2.2=.82	
	30[1]	2103			16 mos. 28/36=.78
Erythromycin	2.5	2258	2 wks. 2.9/3.3=.88	8 mos. 2.3/3.2=.72	
	2.0[1]	2016			14 mos. 2.6/3.3=.79
Gentamicin	9	2255	3 wks. 9.3/8.5=1.09	9 mos. 7.6/8.5=.89	
	10[1]	2101			18 mos. 10.5/10.8=.9
Kanamycin	18*	2261	3 wks. 17/19=.90	11 m.s. 20/20=1.0	
	30[1]	2013			14 mos. 31/36=.86
Methicillin	5^	2260	2 wks. 5.9/7.1=.83	10 mos. 4.5/6.1=.74	
		2043			13 mos. 31/36=.83
Nalidixic Acid	15	2288	3 wks. 18.5/18.4=1.0	8 mos. 17.2/18.3=.94	
	30[1]	1887			14 mos. 45/42=1.07
Neomycin	10*	2279	2 wks. 6.4/9.5=.68		
	20*	2674		3 mos. 15.7/26=.[illegible]8	
	30[1]	1896			23 mos. 25/46=.63
Nitrofurantoin	25*	2287	1 mo. 32/30=1.07		8 mos. 27/30=.90
	15	2534		7 mos. 19.2/21=.91(3)	

Effect of Disk Age on Elution to Diffusion Assay Ratio
(Based on 10 minute Elution Assay Value only)

Antibiotic	Manufactured Nominal Potency, mcg.	Lot No.	Approximate Age at Assay and Elution/Diffusion Ratio		
Novobiocin[1]	2.5*	2269	2 wks. 0.6/1.8=.33	11 mos. 0.4/1.4=.28	
	30†	1876			25 mos. 22/37=.59
Oleandomycin	7.5*	2256	4 wks. 10.2/9.2=1.11		12 mos. 9.9/8.2=1.20
	15†	2107		11 mos. 16.7/16.7=1.00	
Penicillin	0.2	2467	1 mo. .25/.28=.89		
		2368		11 mos. .18/.26=.69	
	0.4*	2267	2 wks. .39/.52=.75	3 mos. .18/.24=.67	17 mos. .29/.41=.71
	10†	2143			12 mos. 8.8/10.1=.88
Polymyxin B	12.5	2482	2 mos. 3.5/16=.22 (0.63)[2]		
		2285		10 mos. 3.1/13.9=.22 (0.64)[2]	
Streptomycin	10†	2280	4 wks. 8.3/11.4=.73	11 mos. 9.5/12.6=.75	
		2100			12 mos. 8.8/11.6=.76
	20	2679	3 mos. 14.8/21.1=.70		
Tetracycline	1.2	2278	2 wks. 1.05/1.38=.76	9 mos. .75/1.17=.64	
	3†	2105			13 mos. 3.7/5.1=.72
	0.5	2462	1 mo. .42/.67=.63		
		2373		9 mos. .37/.60=.62	

TABLE 9

Effect of Disk Age on Elution to Diffusion Assay Ratio
(Based on 10 minute Elution Assay Value only)

Antibiotic	Manufactured Nominal Potency, mcg.	Lot No.	Approximate Age at Assay and Elution/Diffusion = Ratio		
Vancomycin	10	2276	3 wks. 13.7/9.8=1.39	10 mos. 9.3/? 9=.94	
	30	1838			25 mos. 51/37=1.38

(1) In a separate study 10 mcg. handmade disks were eluted 18 hours and 3 months after manufacture for 30 minutes (not 10 minutes), yielding 0.87 and 0.85 of the applied antibiotic, assayed by a spectrophotometric procedure.

(2) The bracketed value expresses elution/diffusion ratios after 3 hours elution, not 10 minutes.

(3) This value expresses elution/diffusion ratio after 30 minutes elution, not 10 minutes.

* Differs from current nominal potency of elution disk.

i Nominal potency of diffusion disk.

A Nominal potency of elution disk and diffusion disk are identical.

TABLE 7

Effect of Shaking on Elution of Nalidixic Acid from 15 mcg. Elution Disks In the Autobac 1 System

Time, min.	Amount eluted, mcg. Shaken(1)	Unshaken(2)
3	8.5	7.2
10	11.2	8.9
15	--	11.6
20	16.3	12.5
30	--	13.7
60	16.2	15.6
120	--	15.7
180	--	15.9

(1) At 36°, 220 oscillations/min. in Autobac 1 cuvette and incubator-shaker with 1.54 ml of phosphate-buffered saline, 270 milliosmolar, pH 7.0, eluant per cuvette chamber. Disk lot 2288.

(2) Exactly as above, but no shaking.

TABLE 8

Micrograms Nalidixic Acid Eluted from 1, 2, 3 Disks per Chamber(1)

Time, min.	1 disk/chamber	2 disks/chamber(2)	3 disks/chamber(2
10	10.5	9.9	8.4
20	15.3	13.3	12.2
30	14.7	11.1	10.5

(1) At 36°, 220 oscillations/min. in Autobac 1 cuvette and incubator-shaker with 1.54 ml phosphate-buffered saline, 270 milliosmolar, pH 7.0, eluant per cuvette chamber. Disk lot 2288.

(2) This simulates elution from a thicker disk; our disk dispenser design will not permit delivery of multiple disks to a single chamber.

TABLE 7

Summary of Optimum Elution Disk Potencies for Use with Autobac I, and Optimum Potency Ranges

Antimicrobial Agent	For Bacterial Class as Identified by Gram Stain	Nominal Potency	Optimal Potency Range, % of Nominal
Ampicillin*	-	3.6 mcg.	80-125%
Ampicillin	+	0.22 mcg.	80-130
Bacitracin	+	18 U.	80-150
Carbenicillin*	-	120 mcg.	80-150
Cephalothin*	+,-	15 mcg.	68-180
Chloramphenicol*	-	4 mcg.	80-150
Clindamycin*	+	2 mcg.	68-150
Colistin*	-	13 mcg.	68-150
Doxycycline	-	1.6 mcg.	80-130
Doxycycline	+	0.5 mcg.	68-180
Erythromycin*	+	2.5 mcg.	68-150
Gentamicin*	+,-	9 mcg.	68-180
Kanamycin*	-	22 mcg.	80-150
Lincomycin	+	2.4 mcg.	68-150
Methicillin*	+	5 mcg.	68-180
Nalidixic Acid	-	15 mcg.	68-180
Neomycin*	-	24 mcg.	68-150
Nitrofurantoin	-	15 mcg.	30-125
Novobiocin	+	2.5 mcg.	68-180
Oleandomycin	+	6 mcg.	68-150
Penicillin G*	+	0.2 U.	68-180
Polymyxin B*	-	12.5 U.	68-150
Streptomycin	-	20 mcg.	80-150
Tetracycline*	-	1.2 mcg.	80-130
Tetracycline*	+	0.5 mcg.	68-150
Vancomycin*	+	10 mcg.	68-180

*Disks for which approval is being sought in this application. Data on other certifiable disks is tentative, and provided for general information only at this time.

See Table 10 for a separate list of disks for which approval is being sought by this application.

Table I

Antibiotic Elution Disks and Permissable Potency Ranges
for which approval is being sought in this application

Disk	Label Potency mcg., (U.)	Permissable Range % of Label
Ampicillin	3.6	80-125%
Carbenicillin	120	80-150
Cephalothin	15	68-180
Chloramphenicol	4	80-150
Clindamycin	2	68-150
Colistin	13	68-150
Erythromycin	2.5	68-150
Gentamicin	9	68-180
Kanamycin	22	80-150
Methicillin	5	68-180
Penicillin G	0.2U.	68-180
Polymyxin B	12.5U.	68-150
Tetracycline (G+ organisms)	0.5	68-150
Tetracycline (G- organisms)	1.2	80-130
Vancomycin	10	68-180

TABLE 11

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg./U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form Numbe
	Ampicillin	2		18		60-97
		10		18		
			.22		18	
--->			3.6*		18	
	Bacitracin	2U.		60		60-97
		10U.				
			18		60	
	Carbenicillin	50		24		61-44
--->			120*		24	
	Cephalothin	30		24		60-97
--->			15*		18	
	Chloramphenicol	5		24		60-97
		30				
--->			4*		24	
	Clindamycin	2		24		61-33
--->			2*		24	
	Colistin	2		24		60-98
		10				
--->			13*		24	
	Doxycycline	5		36		60-98
		30				
			.5		12	
			1.6		12	

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TABLE II (Con't.)

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form 6 Number
	Erythromycin	2		24		60-985
--->		15	2.5*		24	
	Gentamicin	10		18		60-912
--->			9*		18	
	Kanamycin	5		24		60-986
		30				
--->			22*		24	
	Lincomycin	2	2.4	48	48	60-987
	Methicillin	5		18		60-989
--->			5*		18	
	Nalidixic Acid	5		N.A. (36)		-
		30		N.A. (36)		
			15		36	
	Nitrofurantoin	100		N.A. (36)		-
		300		N.A. (36)		
			15		to be estab.	
	Neomycin	5		30		60-991
		30			30	
			24			
	Novobiocin	5		36		60-992
		30			12	
			2.5			
	Oleandomycin	2		24		60-994
		15				

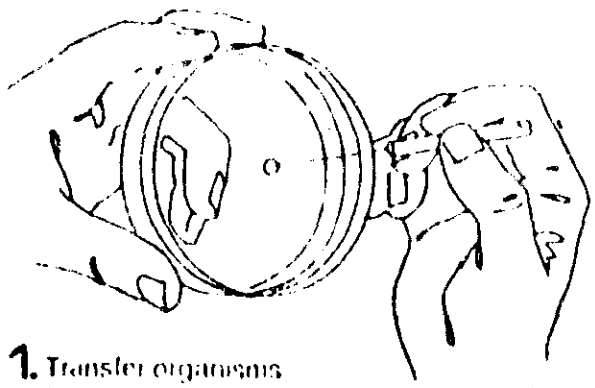
TABLE 11 (Con't.)

Comparison of Diffusion and Elution Disk
Potencies and Claimed Expiration Dates

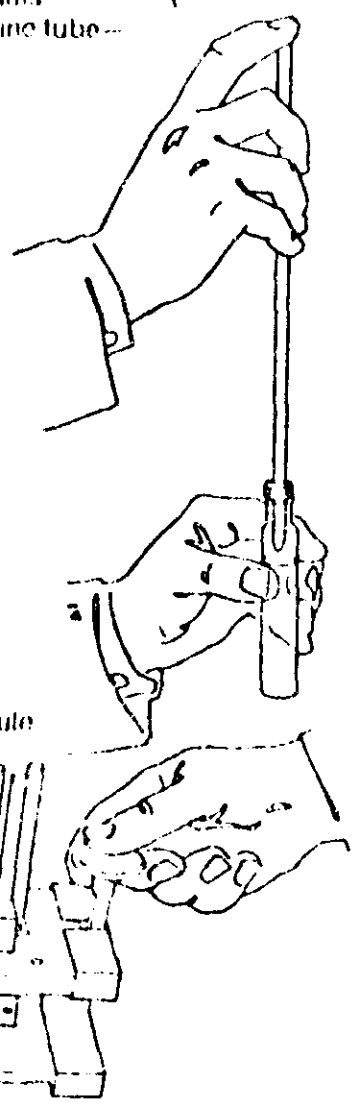
	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form 6 Number
	Penicillin G	2U. 10U.		12		60-997
--->			0.2 U*		12	
	Polymyxin B	50U. 300U.		36		60-999
--->			12.5 U*		to be estab.	
	Streptomycin	2 10		24		61-000
			20		24	
	Tetracycline	5 30		24		61-002
--->			0.5*		12	
--->			1.2*		18	
	Vancomycin	5 30		36		61-003
--->			10*		36	

---> * Approval is being sought for these disks by this application.

As easy as 1, 2, 3 – 4, 5, 6

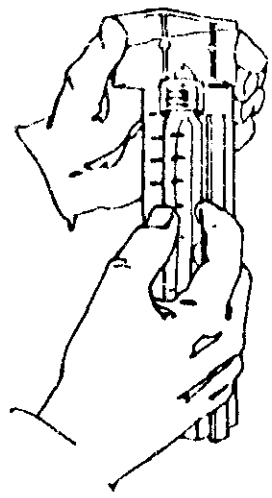


1. Transfer organisms from plate to saline tube – adjust turbidity

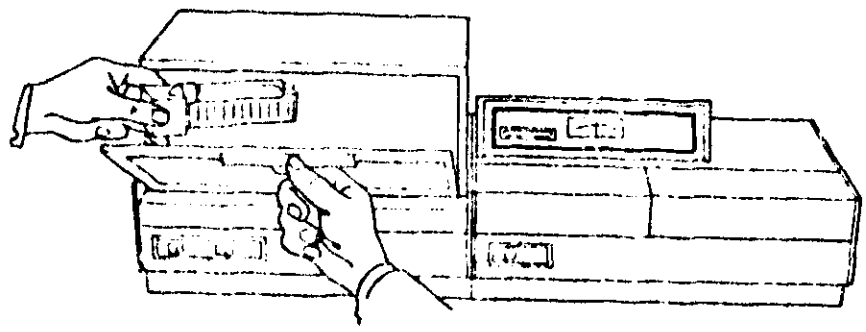


2. Add aliquot to broth tube.

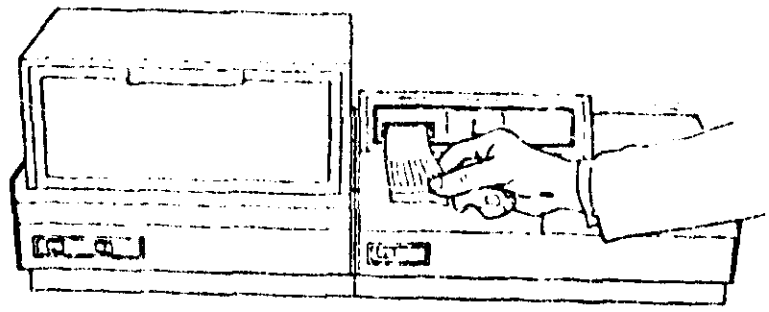
3. Dispense disks into cuvette module



4. Attach inoculated broth tube to cuvette module.



5. Incubate.



6. Read printout.

Automated antimicrobial susceptibility test system

Susceptibility test results in approximately three hours

DIAGNOSTICS DIVISION

APPENDIX 2

Measurement of Rates of Elution of Labeled Tetracyclines from Paper Sensitivity Discs into Eugonic Broth Medium

Joseph F. Dooley

October 25, 1972

The Auto/Bac I antibiotic susceptibility system determines the sensitivity of microorganisms to antibiotics by utilizing impregnated paper discs to introduce specific quantities of antibiotics into a 13 chambered plastic cuvette containing standard bacterial inocula. The concentration of antibiotic within each chamber during the initial 3 hour incubation period, determines, within quite narrow limits, the success of the Auto/Bac I interpretive designation. This study was undertaken in order to determine the rate and extent of elution of tetracycline antibiotics from paper sensitivity discs into eugonic broth growth medium using the conditions of the Auto/Bac I susceptibility system.

Materials and Methods

Tritiated doxycycline[1] hydrochloride hemihydrate hemiethanolate obtained from Pfizer, Inc. was prepared from methacycline hydrochloride by catalytic reduction in the presence of tritium gas.(1) Tritiated tetracycline hydrochloride was obtained from Amersham/Searle Corp. Toluene scintillator contained PPO (4 gm) and POPOP (300 mg) per liter of toluene with 50% (v/v) of Triton X-100 and 1% water and 0.1% formic acid. Scintillation counting was done in a Mark I Nuclear Chicago instrument. The counting efficiency was determined by internal standardization using tritiated toluene (31,400 dpm). Samples were counted for 20 minutes, with counts accumulated in the range of 100,000 - 200,000 for shaking experiments, and 20,000 - 100,000 for non-shaking experiments with counting efficiencies of 14-18%.

Labeled antibiotics were made up to standard solution of known concentration and placed onto paper discs in 10 λ aliquots. After screen drying overnight at room temperature, the discs were dispensed into an Auto/Bac I B chambered cuvette containing 1.54 ml of broth solution (18 ml of eugonic broth/2 ml of saline) in each chamber. The cuvettes were incubated at 37° and shaken in a modified G-25 New Brunswick Shaker/Incubator at a rotational frequency of 220 rpm (3/4" amplitude).

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1. Vibramycin^{R}, α-6-deoxy-5-hydroxy-tetracycline.

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F. A. HOCHSTEIN

Results

The results of these expetiments are shown in Figures 1 and 2. Doxycyline elutes from paper elution discs containing 2.0 mcg into protein broth medium quite rapidly, attaining 82.8% completion in the first ten minutes (Table I). Essentially all (95.9%) doxycycline is eluted from the disc at 1.0 hour. The unshaken reference discs show significantly slower elution, 25.7% after 10 minutes and show a longer elution profile throughout the 3.0 hour experiment. Doxycycline is not completely eluted from the paper, discs after 3.0 hours without shaking. The precision (C.V.) of replicate analysis was 5.5% for shaking and 9.3% for the non-shaking control.

Table I

Percent elution of labeled tetracyclines into eugonic broth/saline solution from paper elution discs at 36°C.

TIME (MIN.)	% DX ELUTED SHAKING	% DX ELUTED NO SHAKING	% TE ELUTED SHAKING	% TE ELUTED NO SHAKING
1.0	44.5	5.3	31.3	31.1
3	67.3	5.8	60.0	32.5
10.0	82.8	25.7	98.2	65.0
20	88.0	47.2	105.0	86.0
40	97.8	62.6	107.6	87.0
60.0	95.9	63.1	107.5	103.7
180.0	97.9	89.1	110.8	106.3

A similar result was found with tetracycline discs containing 1.04 mcg of antibiotic (Table II). A plateau level of 70,508 dpm (1.09 mcg) was observed after 20 minutes of shaking, compared to the control of 57,759 dpm (0.89 mcg). Combustion analysis of 11 tetracycline discs showed an average 67,167 dpm equivalent to a 1.04 mcg disc loading with an S.D. of $\pm$ 1,968 dpm (2.93%). Elution of tetracycline from the discs was more rapid in the non-shaking control than for doxycycline, where complete elution occurred at 60 minutes.

Table II

Elution of ^{3}H-Tetracycline Disc (1.0 mcg)
into Eugonic Broth at 36°C.

TIME INTERVAL (MINUTES)	CONTINUOUS ROTARY SHAKING DPM	% ELUTED	MCG ELUTED	UNAGITATED DPM	% ELUTED	MCG ELUTED
0.5	21,884	32.58	0.34	12,426	18.50	0.19
1.0	21,046	31.33	0.33	20,895	31.11	0.32
2.0	31,474	46.86	0.49	21,995	32.75	0.34
3.0	40,094	56.69	0.62	21,844	32.52	0.34
4.0	46,696	69.52	0.72	24.085	35.86	0.37
5.0	50,814	75.65	0.79	31,353	46.68	0.49
10	65,834	98.02	1.02	43,637	64.97	0.68
15	67,217	100.07	1.04	52,974	78.87	0.82
20	70,508	104.97	1.09	57,759	85.99	0.89
40	72,254	107.57	1.12	58,445	87.02	0.90
60	72,234	107.54	1.12	69.680	103.74	1.08
180	74.394	110.76	1.15	71,427	106.34	1.11

Discussion

Elution of antibiotics from paper discs into growth medium varies as a function of time. Changes in chamber concentration occur rapidly within the first minutes of contact of the disc with protein broth solution. Thus, it becomes relevant to assay protein broth solutions with a precision which is unattainable in the standard microbiological cup plate procedures. These results demonstrate that radioassay techniques using labeled tetracyclines afford a precision (± 5.5%) which allows small changes of antibiotic concentration with time to be observed. Furthermore, this method affords direct assay of disc antibiotic mass levels, which are not available by other techniques.

The results described for doxycycline and tetracycline demonstrate that for these antibiotics rapid continuous rotary shaking is necessary to attain complete solution in short time periods necessary for 3 hour susceptibility testing.

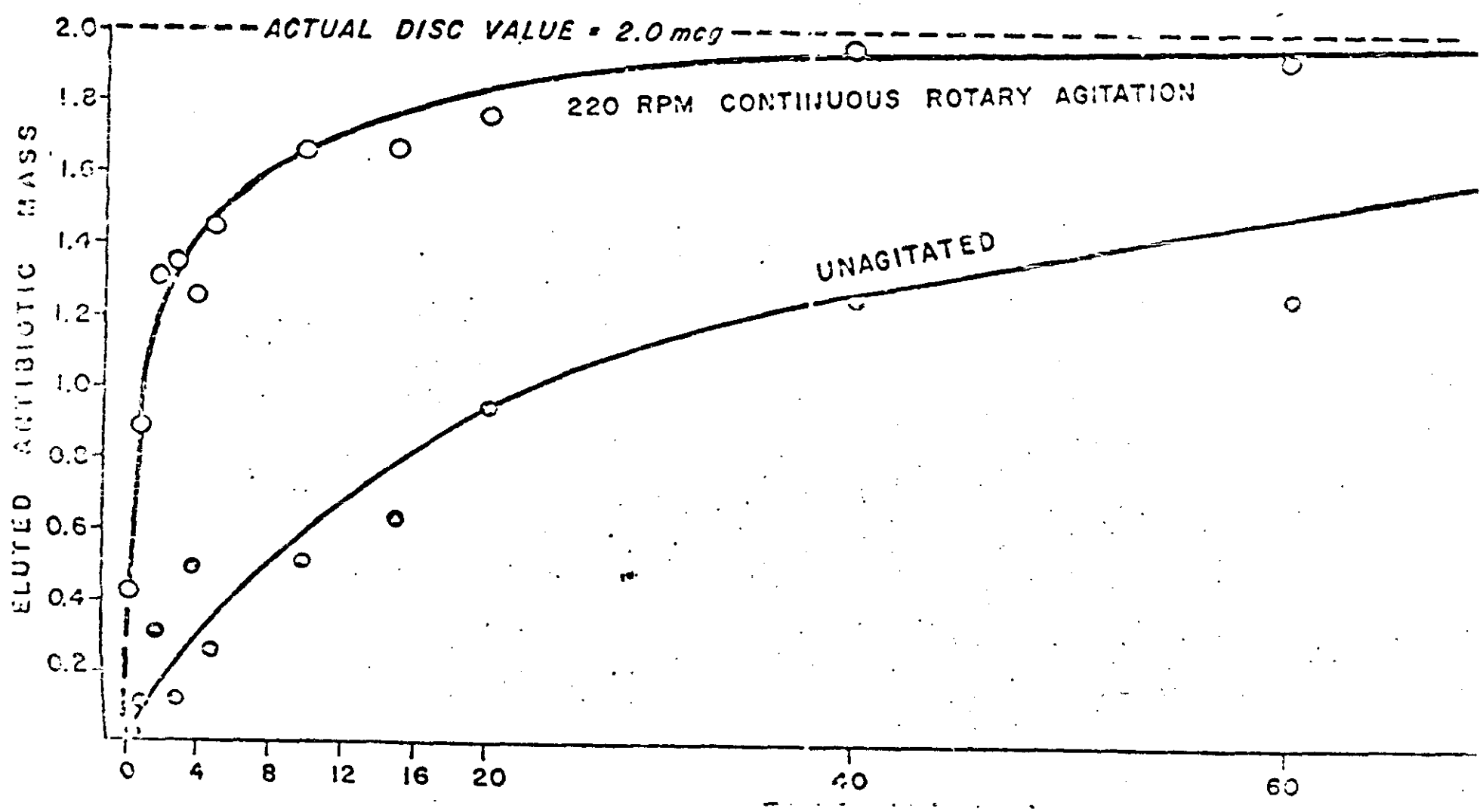
References

1. Stevens, C. R., Beerebbom, J. J., Rennhard, H., Gordon P. N., Murai, K., Blackwood, R. K., and Schach von Wittenau, M., J. Amer. Chem. Soc., 85, 2643 (1963).

Figure 3

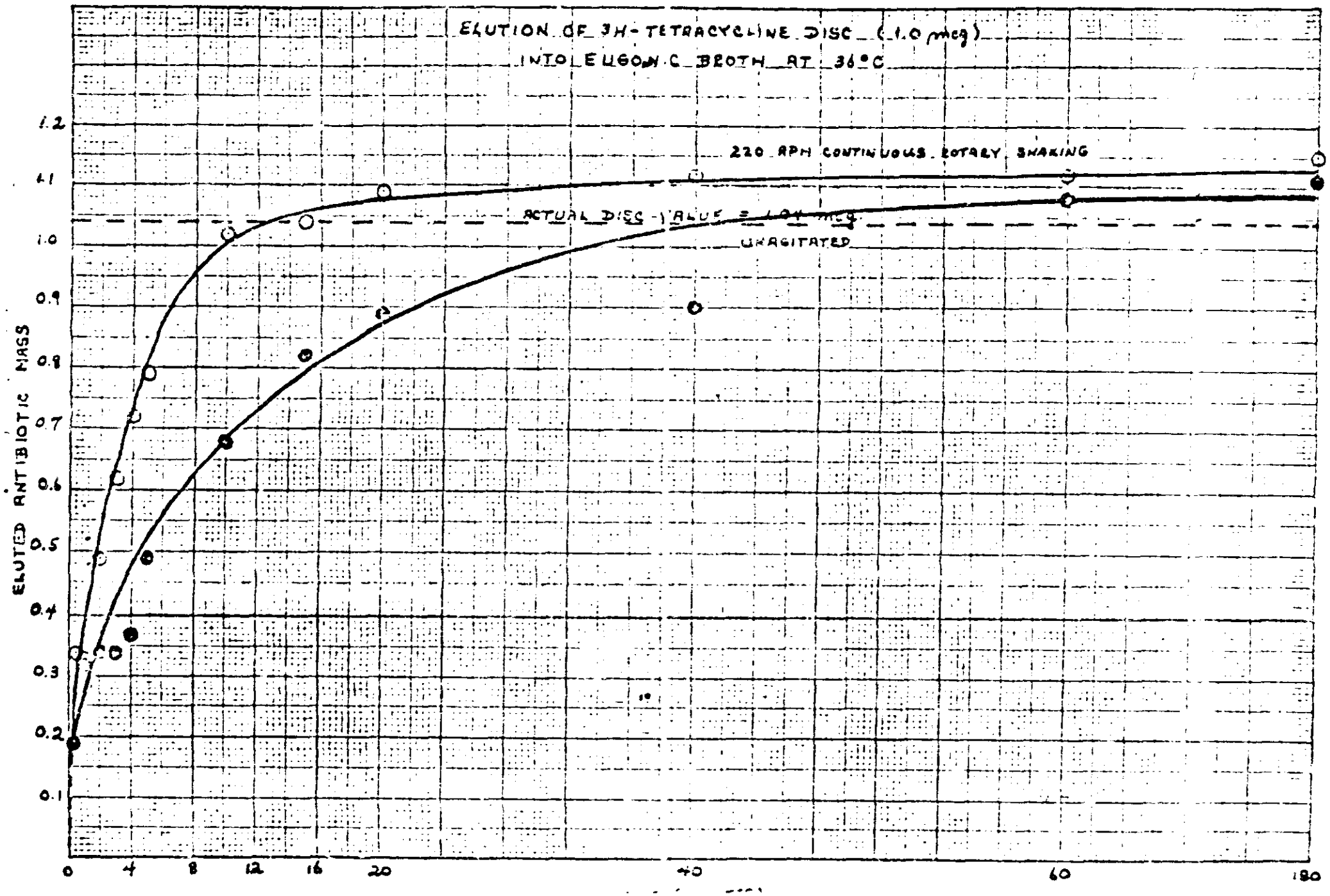
DOXYCYCLINE

ELUTION OF ^{3}H-DOXYCYCLINE DISC (2.0 mcg)
INTO EUGONIC BROTH AT 36°C



K&E 20 X 20 TO THE INCH 46 1240
7 X 10 INCHES MADE IN U.S.A.
KEUFFEL & ESSER CO

TETRACYCLINE



ANCILLARY DATA

The information provided hereafter in Section 4 prior to Section 4a is offered for general background purposes only. The items discussed herein are not certifiable, nor subject to licensing.

Pfizer — STANDARD TEST PROCEDURE

A Spectrophotometric Procedure for Identification and Assay of Nitrofurantoin in 15 mcg. Disks for the Autobac 1

DATE	STP NUMBER
6/73	
PAGE 1 of 2	SUPERSEDES ORIGINAL
STP CITED	

Principle:

Nitrofurantoin is eluted from disks by a buffered eluant, identified by its U.V. absorption curve, and assayed by the intensity of its absorbance peak at 375 nm.[1]

Procedure:

1. Select 10 disks from two different cartridges of the sample, choosing five from the top half of one vial, five from the bottom half of the second vial.

2. Prepare a buffered eluant by adding 500 ml of 0.1 molar monopotassium phosphate, KH_2PO_4 solution (13.6 g/l) to 296.3 ml of 0.1 N NaOH, diluting to 1 liter and adjusting to pH 7 with monopotassium phosphate or sodium hydroxide, if necessary. Then adjust the osmolarity to 270 Mosm/kg by addition of 5 gm NaCl to approximate the osmolarity of Eugonic broth.[2]

3. Place 1 Nitrofurantoin disk in each of ten suitable test tubes, e.g. 12 x 120 mm, add 5.0 ml of buffer solution and stopper with a clean stopper or screw cap.

4. Place on a shaker, e.g. an Autobac 1 shaker or Eberbach shaker, and shake at about 200 oscillations per minute for 30-60 minutes.[3]

5. Remove the test tube stoppers, centrifuge the tubes if necessary to compact paper fibers; decant the clear eluant from each tube successively into a U.V. transparent 1 cm cell.[4]

6. Scan one sample from 420-220 nm in a suitable spectrophotometer, e.g. a Beckman Model DB, vs. a buffer (or water) blank. The curve should show the characteristic Nitrofurantoin peaks at 375 and 270 nm with a minimum at 308 nm, as exemplified by the attached curve, except that absorbtion may be higher at 260-220 nm due to background extracted from the paper.

7. Then read the optical density of the 375 nm peak for each of the remaining nine eluates.

8. Calculate the concentration of Nitrofurantoin in each disk by the following formula.[5]

$$\text{FD content, mcg/disk} = \frac{\text{O.D.}_{375\text{ nm}} \times 5}{.392} \times 5$$

The average Nitrofurantoin content of the ten disks must be in the range 12.7-18.75 mcg/disk. If any single disk shows an assay less than 12.7 or more than 18.75 mcg., an additional ten disks from the same cartridges should be assayed. Not more than two disks in 20 shall be outside this range, and none shall be outside the range 11.5-22.5 mcg/disk.

NOTES:

1. This method is a variant of that described in USP XVIII, pages 448-449.

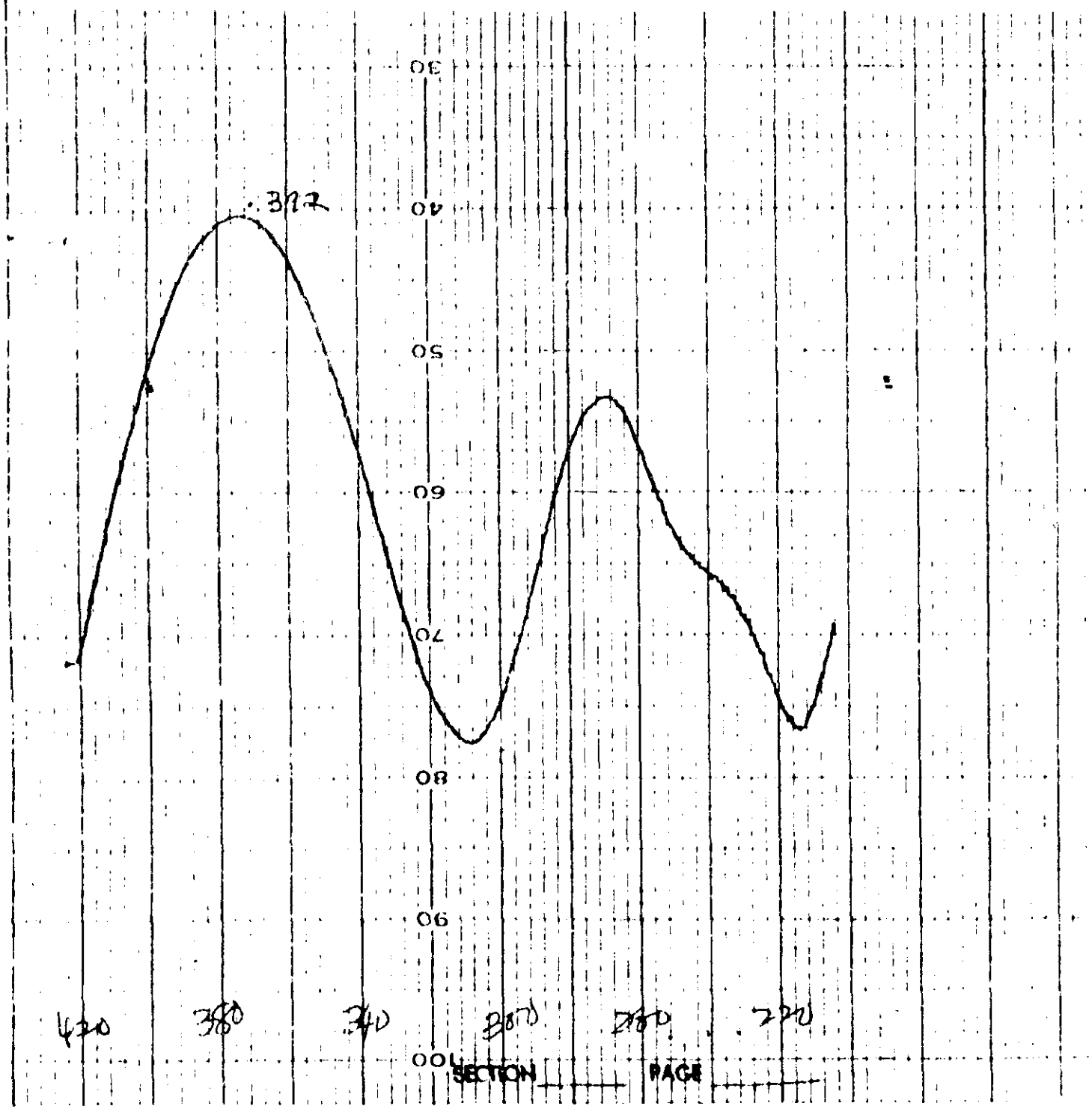
2. This eluate simulates Eugonic broth in pH and osmolarity, but shows no interfering U.V. absorbtion. The U.V. absorbtion spectrum of Nitrofurantoin is not significantly changed over the pH range 5 to 9.

3. Shorter elution times may give incomplete extraction of the Nitrofurantoin. Up to 1-3% more elution may be detected if shaking is extended for an additional 30 minutes.

4. Do not centrifuge more than 5 minutes; solvent is lost on prolonged centrifugation.

5. Pure Nitrofurantoin (USP Standard Lot #G1, assigned potency 1000 mcg/mg) shows an optical density in pH 7 buffer at 375 nm of 0.392 for a 5.0 mcg/ml solution measured in a 1 cm cell.

Ultraviolet absorption curve of Nitrofurantoin, USP Standard Lot G1, 1000 mcg/mg, in 0.1 M phosphate buffer, pH 7.0, adjusted to 270 milliosmolar with sodium chloride. 1.0 cm cell, 5.0 mcg/ml.



STANDARD TEST PROCEDURE

A Spectrophotometric Procedure for Identification and Assay of Nalidixic Acid in 15 mcg. Disks for the Autobac 1

DATE 6/73 | STP NUMBER

PAGE 1 of 2 | SUPERSEDES ORIGINAL

STP CITED

Principle:

Nalidixic Acid is eluted from disks by a buffered eluant, identified by its U.V. absorption curve, and assayed by the intensity of its absorbance peak at 330 nm.[1]

Procedure:

1. Select 10 disks from two different cartridges of the sample, choosing five from the top half of one vial and five from the bottom half of the second vial.
2. Prepare a buffered eluant by adding 500 ml of 0.1 molar potassium phosphate, KH_2PO_4 solution, to 296.3 ml of 0.1 N NaOH, diluting to 1000 ml with distilled water and adjusting the pH to 7.0 with monopotassium phosphate or sodium hydroxide if necessary. Then adjust the osmolarity to 270 Mosm/kg by addition of 5 gm NaCl.[2]
3. Place 1 Nalidixic Acid disk in each of 10 suitable test tubes, e.g. 12 x 120 mm, and add 5 ml of buffer to each tube. Stopper with a clean stopper or screw cap.
4. Place on a suitable shaker, e.g. an Autobac 1 or Eberbach, and shake for 30-60 minutes; centrifuge if necessary to compact the paper[4] and decant the clear eluate from each tube into a 1 cm U.V. transparent cell.
5. Scan one sample from 350-220 nm vs. a buffer blank in a suitable spectrophotometer, e.g. a Beckman Model DB. The curve should show the characteristic Nalidixic Acid peaks at 330 and 254 nm with minima at 274 and 234 nm (as exemplified by the attached curve), except that absorption may be higher at 260-220 nm due to background extracted from the paper.
6. Read the optical density of the 330 nm peak for each of the nine remaining eluates.
7. Calculate the concentration of Nalidixic Acid in each disk by the following formula:[3]

$$\text{Nalidixic Acid Content (mcg/disk)} = \frac{\text{O.D.}_{330\ nm} \times 10}{.420} \times 5$$

The average Nalidixic Acid content of the ten disks must be in the range 10 to 27 mcg/disk. If any single disk shows an assay outside this range, an additional ten disks from the same cartridge should be assayed. No more than 2 disks in 20 shall be outside this range, and none shall be outside the range 8 to 30 mcg.

NOTES:

1. This method is a variant of that described in N.F. XIII, page 466.
2. This eluate simulates Eugonic broth in pH and ionic strength, but shows no interfering U.V. absorption. The U.V. absorption spectrum of Nalidixic Acid is not significantly changed over the pH range 5.7 to 9.5.
3. Pure Nalidixic Acid (Sterling-Winthrop, Lot #R-002-XA) shows an optical density in pH 7 buffer at 330 nm of .420 for a 10.0 mcg/ml solution measured in a 1 cm cell. This Nalidixic Acid had an assigned potency of 1000 mcg/mg.
4. Do not centrifuge more than 5 minutes, to avoid excessive solvent loss.

5. Nalidixic Acid, Lot #R-002-XA from Sterling-Winthrop, with a nominal potency of 1,000 mcg/mg, was used to establish a Nalidixic Acid Standard Curve. 5.0 mg of Nalidixic Acid was dissolved in pH 7.0 KH_2PO_4 buffer and Q.S. to 100 ml. (In order to dissolve all of the Nalidixic Acid, the solution had to be stirred on a magnetic stirrer for approximately 1 hour.) This solution was then diluted 1:10, 2:10, 3:10 and 5:10 to yield solutions containing 5.0, 10.0, 15.0 and 25.0 mcg N.A./ml, respectively. The O.D. of the solutions was measured by "searching for the peak" at approximately 330 nm and recording the maximum O.D. (Beckman DB).

Solution	O.D. @ 330 nm	C.F. ($\frac{conc.}{O.D.}$)
5 mcg/ml	.215	23.26
10 mcg/ml	.420	23.81
15 mcg/ml	.624	24.04
25 mcg/ml	1.046	23.90

average C.F. = 23.75 (curve attached)

Investigation into the effect of pH changes on Nalidixic Acid:

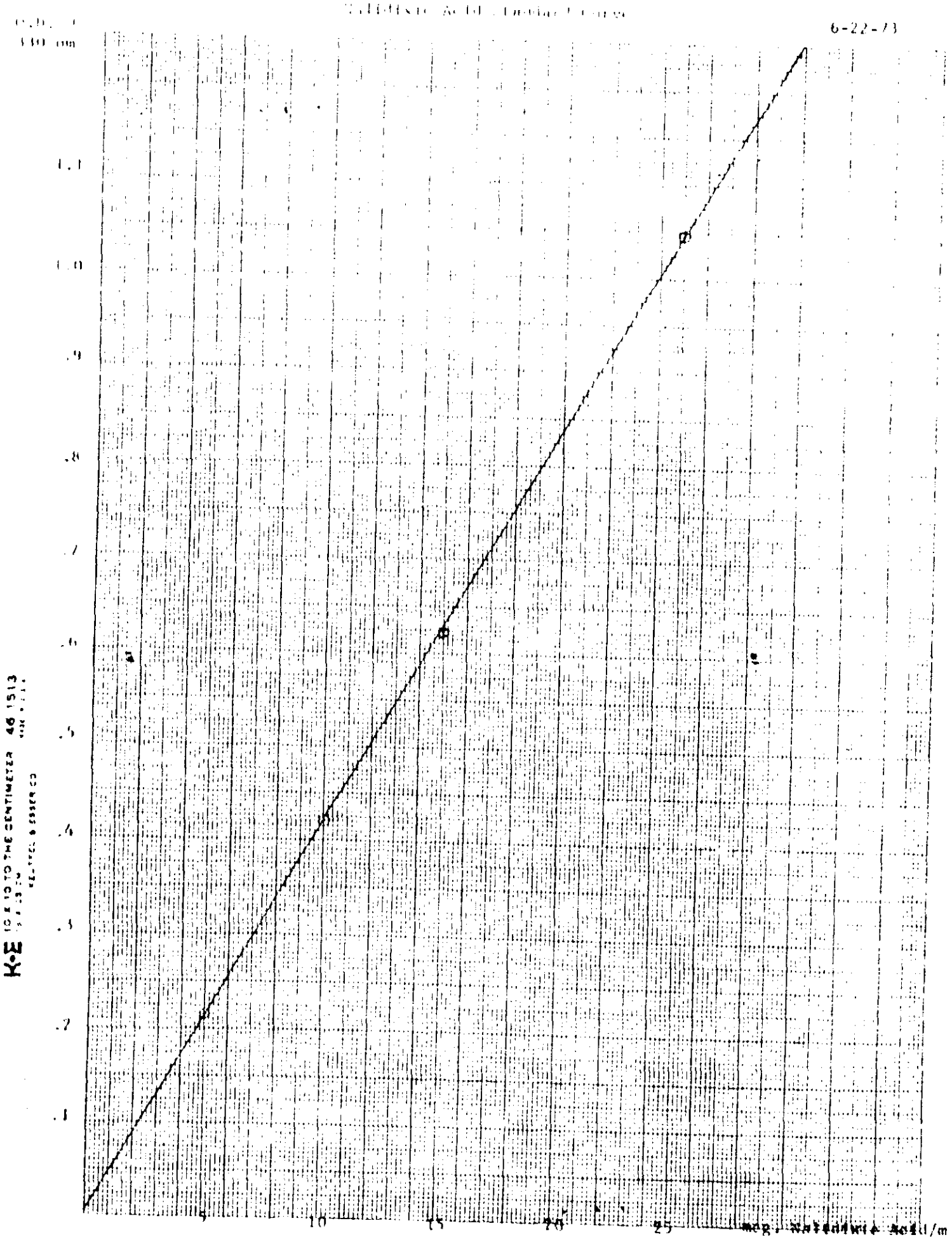
Solutions of Nalidixic Acid were made up and the pH adjusted to range from 4.5-9.5. The positions of the peaks were compared to the solution at pH 7.0. (Gulford Spectrophotometer)

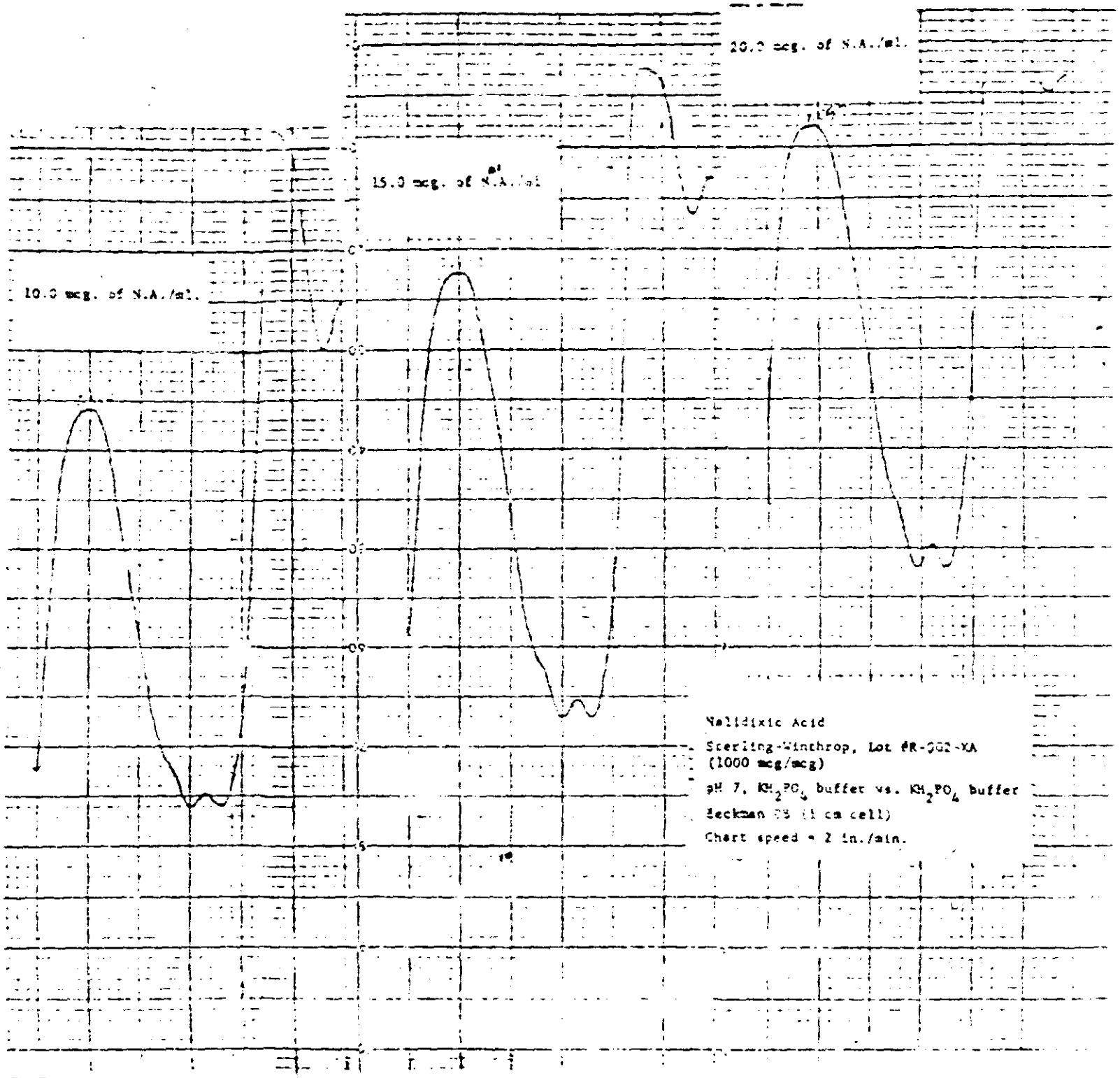
pH	peak location
9.5	257 nm and 334 nm
7.0	257 nm and 332 nm
5.7	257 nm and 328 nm
4.5	247 nm and 306 nm

This data indicates that the peak locations are not significantly altered in the pH range of 5.7-9.5.

C.V. of the lots tested in December, 1972 was calculated for the data collected at 10, 30, 60 and 180 minutes:

Lot #	C.V.	n
2425	0.096	11
2426	0.056	12
2288	0.084	12


6-22-73
1.1
1.0
.9
.8
.7
.6
.5
.4
.3
.2
.1
5
10
15
20
25
mg. Nitrilotriacetic Acid/m


10.0 mcg. of N.A./ml.
15.0 mcg. of N.A./ml.
20.0 mcg. of N.A./ml.
Nalidixic Acid
Sterling-Winthrop, Lot #R-002-XA
(1000 mcg/mcg)
pH 7, KH_2PO_4 buffer vs. KH_2PO_4 buffer
Beckman DB (1 cm cell)
Chart speed = 2 in./min.

STANDARD TEST PROCEDURE

A Microbiological Assay Procedure for the Assay of Nitrofurantoin and Nalidixic Acid in 15 mcg. Elution Disks for the Autobac 1	DATE 6/73 PAGE 1 of 1 STP CITED	STP NUMBER SUPERSEDES ORIGINAL

Principle:

The procedure is identical to the general procedure described for certifiable antibiotics in CFR 147.1.

Procedure:

Proceed exactly as directed in CFR 147.1 for the assay of antibiotic susceptibility discs, with the following inoculum, media and incubation conditions.

	Nitrofurantoin	Nalidixic Acid
Base Layer	Mueller Hinton, 42 ml	Antibiotic Medium No. 2, 42 ml
Seed Layer	Mueller Hinton, 8 ml	Antibiotic Medium No. 1, 8 ml
Organism, volume	E. coli, 1.5 ml	E. coli, 1 ml
Inoculum density*	80% T	80% T
Incubation Temp.	33.5°	35°
Standard Curve points, mcg.	3, 6, 12, 24, 48	3, 6, 12, 24 & 48

*Measured at 650 nm in a 16 mm tube.

Calculate the results as described in CFR 147.1.

June 16, 1972

Nitrofurantoin and Nalidixic Acid Content of Sensitivity Disks by UV Assay

Nitrofurantoin

Nitrofurantoin (FD), potency 1000 mcg/mg, was used as a standard. A 5.0 mcg/ml solution in KH_2PO_4 buffer was scanned in 1 cm cells vs. buffer on the Beckman, DB spectrophotometer from 460 to 220 nm. The KH_2PO_4 buffer was made by addition of 500 ml 0.1 M KH_2PO_4 to 296.3 ml 0.1 N NaOH then diluting to 1000 ml with distilled water. The pH of the solutions was 7.0. Osmolarity was adjusted to 270 Mosm/kg by addition of 5 gm. NaCl to approximate the osmolarity of Eugon broth (273 Mosm/kg).

Nitrofurantoin disks from Pfizer's experimental lot, No. 2287, with a nominal potency of 25 mcg/disk, and Nitrofurantoin 300 mcg sensitivity disks, Lot No. 2023 (Pfizer), 1185 (Pfizer), 566998 (DIFCO), 106048 (BBL) were tested for Nitrofurantoin by elution and UV assay. For each assay time four disks from each lot were placed in separate chambers of an elution cuvette which was filled with 20 ml of KH_2PO_4 buffer (about 1.5 ml/chamber). The cuvette was then placed in a shaker, and samples of the solution were collected after 4, 10, 20 and 30 minutes of shaking in the Brunswick shaker at 220 oscillations per minute at 37°C. The combined samples from the four disks for each time period were then diluted 1:100 (1:10 dilution for the experimental lot, No. 2287) with distilled water and scanned against water on a Beckman DB spectrophotometer from 460 to 220 nm.

In order to compare the effects of elution by Eugon broth and KH_2PO_4 buffer, the following procedures were followed (Eugon broth is not a suitable solvent for the UV determination of Nitrofurantoin or Nalidixic Acid):

1. Four disks from the experimental lot were eluted in Eugon broth, and four in KH_2PO_4 buffer for 30 minutes.

2. The extracted disks were then blotted with tissue, and re-eluted in KH_2PO_4 buffer for a second 30 minute period.

3. Samples from the first elution of four disks by buffer and from the second elution of four disks by both buffer and by Eugon broth were combined and then scanned on the Beckman DB from 460 to 220 nm to measure the Nitrofurantoin recovered in the second extraction operation.

All scans showedthe characteristic Nitrofurantoin absorption peaks at 375 nm and 270 nm. The results are listed in Table I.

Nalidixic Acid

Nalidixic Acid (NA), potency 1000 mcg/mg, was used as a standard. A 5.39 mcg/ml solution in KH_2PO_4 buffer was scanned in 1 cm cells vs. buffer on the Beckman DB spectrophotometer from 400 to 240 nm. The same buffer was used as in the Nitrofurantoin (FD) assay described above. NA, experimental lot, No. 2288, with a nominal potency of 12.5 mcg/disk, and NA, 300 mcg sensitivity disks Lot 2075 (Pfizer), 1887 (Pfizer), 109065 (BBL) and 571556 (DIFCO) were tested for NA content by elution and UV assay. The test procedures

were the same as in FD assay except that all samples were diluted 1:10 with distilled water. The characteristic NA absorption peaks at 330 nm and 255 nm were seen in all extracted samples. The results are listed in Table II.

R. Yang

RY:hlg

cc: Dr. J. Hackett
Dr. G. Evanega

SECTION 8 PAGE 9

TABLE 1

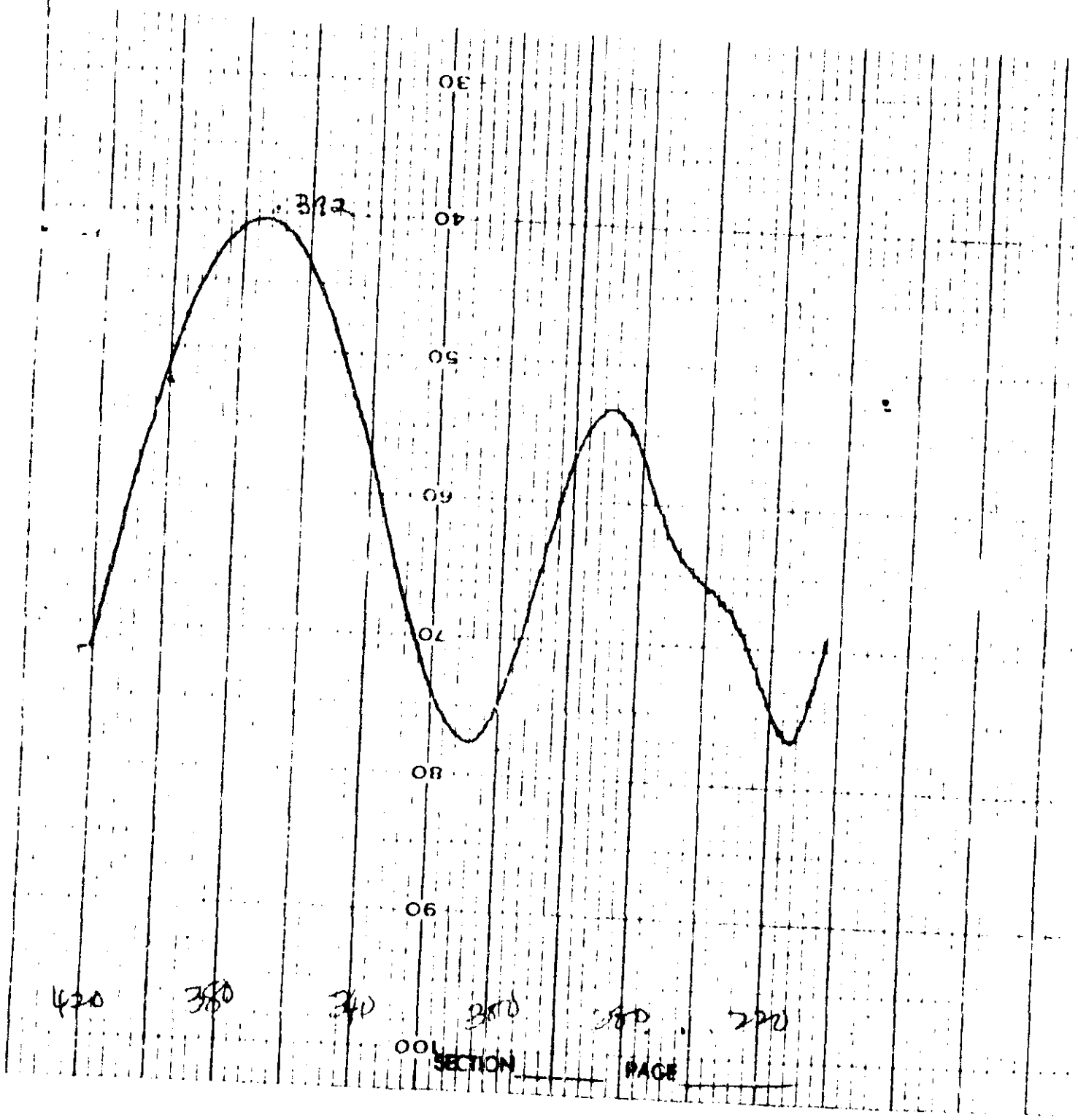
SAMPLE	ELUTION TIME	SCAN NO.	OD at 375 nm	OD at 270 nm	FD CONTENT mcg/dis (based on 375 nm pe
Pfizer Lot 2023, 300 mcg.	30 min.	1	.126	.111	237
Pfizer Lot 1885, 300 mcg.	30 min.	2	.138	.112	260
DIFCO Lot 66998, 300 mcg.	30 min.	3	.108	.078	204
BBL Lot 106048, 300 mcg.	30 min.	4	.118	.085	222
Experimental Lot 2267 nominal potency 25 mg/disk	4 min.	9	.149	.114	28.1
	10 min.	9	.169	.129	31.8
	20 min.	9	.176	.131	33.0
	30 min.	9	.196	.149	36.9
Experimental Lot 2nd elution after Eugon broth extraction	30 min.	9	.034	carry over from Eugon broth	1.1
Experimental Lot 2nd elution after buffer extraction	30 min.	9	.036	.051	1.1

NOTE: The solubility of FD in H_2O at pH 7.0 is 280 mcg/1.5 ml (Merck Index)

TABLE II

SAMPLE	ELUTION TIME	SCAN NO.	OD at 330 nm	NA CONTENT mcg/disk
Pfizer 2075, 30 mcg.	30 min.	5	.143	43.5
Pfizer 1887, 30 mcg.	30 min.	6	.143	43.5
BBL Lot 109065, 30 mcg.	30 min.	7	.149	45.4
DIFCO Lot 571556, 30 mcg.	30 min.	8	.109	33.2
Experimental Lot 2288 nominal potency 15 mcg/disk	4 min.	10	.061	18.5
	10 min.	10	.061	18.5
	20 min.	10	.061	18.5
	30 min.	10	.061	18.5
Experimental Lot 2nd elution after Bugon broth extraction	30 min.	10	.012	0.6
Experimental Lot 2nd elution after buffer extraction	30 min.	10	.012	0.6

Ultraviolet absorption curve of Nitrofurantoin, USP Standard Lot G1, 1000 mcg/mg, in 0.1 M phosphate buffer, pH 7.0, adjusted to 270 milliosmolar with sodium chloride, 1.0 cm cell, 5.0 mcg/ml.



RELEASE - R

10/73	SUPERSEDES 6/73	Eugonic Broth for the Autobac 1 System
1 or 2	CHART	

ackground:

ugonic broth for the Autobac 1 has the following approximate formula:

er liter of distilled water.

ppearance:

pale to medium yellow aqueous solution, free of turbidity or gross particulate matter. Tubes shall be clean and properly closed.

dentity:

ssay:

201-5A (FRONT)
10-72

CONFIDENTIAL

CONFIDENTIAL

001-9A (FRONT) 10-72

RELEASE - R

10/73	SUPERSEDES 6/73	Inoculum Standardization Solution for the Autobac 1
1 of 1	GRADE	

[redacted]

OLI-5A (FRONT) 10-72

CONFIDENTIAL

July 18, 1973

Stability of Eugonic Broth

Eugonic broth, used as a growth supporting medium in the Autobac I system must, to be satisfactory, maintain adequate clarity and sufficient growth supporting ability to allow detection of the inhibition of microbial growth by antimicrobial agents within the normal three hour test cycle. The Eugonic broth discussed herein was manufactured according to the standard formula. Each liter of broth contains approximately 14 grams Casein Hydrolysate, 5 grams Vegetable Protein Hydrolysate, 4 grams Sodium Chloride USP, 0.2 grams Sodium Sulfite, 0.7 grams L - cysteine H Cl, 5.5 grams Dextrose, 1.0 grams Sodium Citrate USP, pH 7.1. The manufacturing cycle differs from that of the standard product only in that a filtration step procedes tube filling and sterilization to ensure a clear product.

The growth supporting ability of Eugonic broth was assessed by inoculating 5.5 ml of medium with 0.1 ml of an overnight agar plate culture of the following microorganisms; after washing the culture off and suspending to a density of 50% T at 530 nm in a 16 mm tube.

Streptococcus Group D, Strain No. En 1
Staphylococcus epidermidis, Strain No. Staph 1
Proteus mirabilis, Strain No. P. mir. 1
Escherichia coli, Strain No. H1
Klebsiella pneumoniae, Strain No. Kl& 2

The strain numbers refer to stock cultures of clinical isolates maintained in our laboratories for control and testing purposes. Growth curves were measured by following turbidity increases at 530 nm in a 16 mm tube over a six hour period. The figures quoted in tabular form below refer to the percent transmittance of the culture at 6 hours. In general, this appears to be a useful reflection of growth supporting ability of the broth. Different organisms show different lag phases, but all have entered well into the log phase at 6 hours. It is recognized that these growth conditions are not identical to those encountered in the Autobac where shaking and excellent aeration is operative, but they do provide a measure of the inherent growth supporting ability of the medium under test.

Data are provided for four lots of Eugonic broth in the attached table. These were stored at 5°, 25° and 35° for periods up to 14 months. The following general conclusions can be drawn.

1) In general, Eugonic broth will develop a deeper yellow color on storage at 35°.

2) The pH of Eugonic broth appears to drop on high temperature storage, never to rise.

3) There is no detectable loss in growth supporting ability for the five organisms tested up to 12 months at 5°. Losses in growth supporting ability after 12 months at 25° are marginal. Losses in growth supporting ability can be detected within 6-7 months at 35°, and become more pronounced after 12 months at 35°.

The correlation between growth supporting data obtained in the above studies and suitability of Eugonic broth for use in the Autobac I were established by examining the growth supporting characteristics of Eugonic broth aged under several conditions in the Autobac. These studies are described in a separate report by J. A. McKie to F. A. Hochstein, June 26, 1973, "Stability Data on Prepared Tubes of Phosphate Buffered Saline and Eugonic Broth for use with the Autobac Method."

<u>Summary</u>

Based upon the above studies and those reported by Dr. McKie in a separate report, Eugonic broth manufactured by Pfizer Diagnostics for use in the Autobac can successfully be used for at least one year after the date of manufacture if stored at approximately 5°C. Storage for one year still permits acceptable growth supporting ability. Prolonged storage at elevated temperatures, e.g. 35°, must be avoided.

J. Hackett, Ph.D.

F. A. Hochstein, Ph.D.

Growth Supports by Eugonic Broth

Lot 23009
Manufactured March, 1972[3]

		pH	Clarity	Color[2]	Growth, % Transmittance at 6 hours[1] Strep.	Staph.	Prot.	E.coli	K.pn.
	Initial	7.1	OK	sy	57±12	81±5	64±12	53±5	55±10
5°	7 mos.	-	OK	sy	76	86	61	53	49
	12 mos.	7.1	OK	sy	43	80	62	48	55
25°	7 mos.	-	OK	sy	74	85	63	54	51
	12 mos.	7.05	OK	py	49	82	67	52	57
35°	7 mos.	-	OK	py	85	88	67	63	53
	12 mos.	6.8	OK	y	80	83	80	59	65

Lot 22267
Manufactured February, 1972[3]

		pH	Clarity	Color[2]	Growth, % Transmittance at 6 hours[1] Strep.	Staph.	Prot.	E.coli	K.pn.
	Initial	7.0	OK	sy	57±12	81±5	64±12	53±5	55±10
5°	7 mos.	-	-	sy	70	85	62	54	48
	12 mos.	7.0	OK	sy	49	76	67	50	58
25°	7 mos.	-	-	sy	74	86	63	56	49
	12 mos.	6.8	OK	py	43	78	70	54	61
35°	7 mos.	-	-	-	85	88	70	62	53
	12 mos.	6.6	OK	y	77	78	81	54	68

Growth Support by Eugonic Broth

Lot 24061
Manufactured April, 1972[3]

		pH	Clarity	Color[2]	Growth, % Transmittance at 6 hours[1] Strep.	Staph.	Prot.	E.coli	K.pn.
	Initial	7.0	OK	sy	57±12	81±5	64±12	53±5	55±10
25°	4 mos.	-	-	-	-	-	71	58	56
	6 mos.	-	-	-	70	76	68	63	58
	14 mos.	6.8	OK	y	74	79	75	60	63

- -

Lot 28155
Manufactured August, 1972[4]

		pH	Clarity	Color[2]	Growth, % Transmittance at 6 hours[1] Strep.	Staph.	Prot.	E.coli	K.pn.
	Initial	7.0	OK	sy	57±12	81±5	64±12	53±5	55±10
5°	6 mos.	-	-	sy	50	80	77	56	65
	9 mos.	6.95	OK	sy	64	77	72	52	67
25°	6 mos.	-	-	sy	45	75	74	67	67
	9 mos.	7.1	OK	py	85	80	77	62	64

(1) Initial values estimated from the average transmittance of 8 lots, stored at 5°, measured within 3 months of preparation.

(2) Color: sy=slightly yellowish py=pale yellow y=yellow

(3) Made from dehydrated medium lot No. 51501

(4) Made from dehydrated medium lot No. 25268

June 26, 1973

TO: Dr. L. A. Hochstein

FROM: J. E. McKie, Jr.

SUBJECT: STABILITY DATA ON PREPARED TUBES OF PHOSPHATE BUFFERED SALINE AND TUGONIC BROTH FOR USE WITH THE AUTOBAC METHOD.

1. PHOSPHATE BUFFERED SALINE. A 0.45 micron membrane-filtered, terminally autoclaved (121°C, 15 minutes) glass tubed and capped product of the following composition per liter of solution: 4.22 grams NaCl, 3.20 grams K_2HPO_4, 1.58 grams KH_2PO_4.

   A. pH Stability of the Final Product at Various Temperatures in Various Glass Tubes.

      The data presented in TABLE I was collected by the following procedure: Product tubes were removed periodically from the temperature chambers (incubators, water baths) and allowed to come to room temperature. They were then shaken several times by inversion, uncapped, and the pH measured using a Beckman Model Expandomatic SS-2 pH meter employing a Corning 476050 semi-micro combination glass electrode. The pH meter was always calibrated with pH 7 reference buffer solution prior to taking sample readings. After reading the pH, the sample was discarded. The various types of glass tubes (16 x 125 mm) used in this study are coded in TABLE I as follows: A = Type I (borosilicate) glass, reusable, Corning Glass; B = Type I (borosilicate) glass, disposable, Corning Glass; C = Type III (soda-lime) glass, disposable, Demuth Div. of Brockway Glass; D = Type I (borosilicate) glass, Demuth Div. of Brockway Glass; E = Type I (borosilicate) glass, Kimble Glass; and F = Type II (soda-lime, fluoride washed) glass, disposable, Demuth Div. of Brockway Glass.

      Analysis of the data shows that the pH of the phosphate buffered saline is unchanged after ca. seven months at 26°C, 37°C, and 45°C when the solution is stored in a sealed Type I glass tube. Furthermore, when the temperature is elevated to 65°C, no change in pH is observed even after ca. 3 months. Assuming that the rate of reaction(s) leading to pH shifts, will double for every 10° rise in temperature, the 45°C stability of the pH, by extrapolation from the 65°C data, should be approximately 3 months x 16 = 4 years for this phosphate-buffered saline solution in the capped borosilicate test tube. The use of Type II and Type III glasses is not routinely recommended on the basis of the pH increases observed, particularly at 37° and 45°C. The maximum time and temperature for storage should be 3 months at 26°C. Ideally, if such glass is to be used, storage at 4°C is recommended.

3. Sterility Stability of the Final Product at Various Temperatures in Various Glass Tubes.

   All tubes of the phosphate-buffered saline for which pH readings

TABLE I

STABILITY OF PUROMYCIN PHOSPHATE-BUFFERED SALINE

Elapsed Storage Time (days)	T = 20 ± 1°C			T = 37 ± 1°C			T = 45 ± 2°C			T = 0° ± 2°C					
	A*	B*	C*	A	B	C	A	B	C	A	B	C	D*	E*	F*
0	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00					
1	7.00	7.00	7.00	7.00	7.00	7.05	7.00	7.00	7.05	7.00	7.00				
2										7.00	7.00				
3										7.00	7.00	7.10			
4										7.00	7.00	7.10	7.00		
6	-	-	-	7.00	7.00	7.05	7.00	7.00	7.00	7.00	7.00	7.20			
7															
11										7.00	7.00	7.20			
13	7.00	7.00	7.05	7.00	7.00	7.09	7.00	7.00	7.12					7.00	7.04
14										7.01	7.01	7.31			
18													7.00		
20	7.00	7.00	7.05	7.00	7.00	7.09	7.00	7.00	7.12						
21															
27	7.00	7.00	7.05	7.00	7.00	7.10	7.00	7.00	7.20					6.97	7.02
30										7.01	7.01	7.63			
31															
32														6.97	7.03
55	7.00	7.00	7.00	7.00	7.00	7.15	7.00	7.00	7.20				7.00		
60															
83	7.02	7.00	7.08	7.02	7.00	7.20	7.02	7.00	7.30					6.97	7.07
88															
216	7.00	7.00	7.10	7.00	7.00	7.29	7.00	7.00	7.88					6.93	7.10

*Various types of 16x125 mm glass test tube containers .... see text for explanation of codes A - F.

(shown in TABLE I) were obtained at 26°, 37°, and 45°C were also sampled for sterility. Sterile BHI broth and sterile BHI agar were inoculated aseptically with the saline and incubated overnight at 37°C. In no case were any viable organisms ever found throughout the 83 day stability period investigated. The terminal autoclaving (121°C, 15 minutes) of the tightly capped tubes ensures a sterile product, and the integrity of the cap seal precludes viable organisms from entering the tubes.

C. Turbidity Stability of the Final Product at Various Temperatures in Various Glass Tubes.

After seven months at 26°C, 37°C, and 45°C, the 35° angle light scattering (function-turbidity) of the phosphate-buffered saline was measured in the sealed product tube and compared with the original light scattering. The same calibrated Autobac photometer was used for these readings in which the tube was placed in the beam in such a manner to prevent scratches on the glass from being in the optical path. No change in the light scattering of the buffered saline was found at any temperature when the storage container was Type I glass. However, the gradual accumulation of a hazy deposit on the inner surface of the Type III glass, particularly evident as the storage temperature increased, prevented reading of the solution light scattering. On vortexing this white surface deposit could be partially transferred to the solution bulk appreciably increasing the scattering. This provides an additional reason why the use of Type III glass is unsatisfactory for long time storage of the buffered saline at room temperature and above. The results are shown in TABLE II below:

TABLE II

Storage Temp. (°C)	Storage Time (months)	Autobac Light Scattering† (see also Section I.C.) Glass A*	Glass B*	Glass C*
-	0	3.60 ± .1	3.60 ± .1	3.60 ± .1
26 ± 1	7	3.66	3.56	3.42
37 ± 1	7	3.68	3.54	hazy
45 ± 2	7	3.58	3.53	very hazy

*see page 1., † $-\log_{10}$ (light scattering intensity at $\leq = 35^{\circ}$) in volts.

II. EUGONIC BROTH. A 0.45 micron membrane - filtered, terminally autoclaved (121°C, 15 minutes) glass tubed and capped product of the following approximate composition in grams per liter of final solution: peptone "C" = 5.0; peptone "S" = 5.0; dextrose = 5.5; sodium chloride = 4.0; sodium sulfite = 0.2; and L- cysteine = 0.7.

A. Growth Support Stability as Used in the Autobac Protocol with Reference Organisms.

Reference microorganisms Escherichia coli ATCC 25922 and Staphylococcus Aureus ATCC 25923 have been used to monitor the short time (3 to 5 hours)

TABLE III

Fluid Broth		Temperature of Fluid Broth Tubes During Storage	Time of Storage	Autoclave Growth Index = Optical Density				
ATCC Lot No.	Prepared Tube Lot No.	(°C)	(months)	ATCC 25922				
				t=3 hrs	4 hrs	5.5 hrs	t=3 hrs	
51601	Research 1	4	0	1.16 ± .20†	1.33	1.65	0.53	
51601	22267	4	9	0.87	1.15	-	0.61	
51601	22267	4	16	1.12	1.37	1.69	0.61	
51601	Research 1	Room Temperature	0	1.16	1.33	1.65	0.52	
51601	22267	Room Temperature	16	0.50	0.99	1.20	0.50	
51601	Research 1	35	0	1.16	1.33	1.65	0.53	
51601	22267	35	9	0.35	0.81	1.21	0.53	
51601	23003	Room Temperature	10	1.11	1.36	1.61	0.51	
---	23291	4	9	1.02	-	-	0.72	
---	23155	4	10	1.05	1.25	1.57	0.58	
---	23155	Room Temperature	10	1.15	1.42	1.70	0.57	
51601	23003 (aseptic)	4	9	0.93	-	-	0.63	
51601	23003 (aseptic)	4	15	0.92	1.27	1.59	0.52	
51601	23003 (aseptic)	Room Temperature	15	0.49	0.92	1.08	0.25	0.41
51601	23003 (aseptic)	35	9	0.33	0.75	0.95	0.21	0.05

† Approximate variation in growth index [illegible] with same lyophil on a [illegible]

growth support at 36°C of the Eugonic Broth as a function of storage time at various temperatures of several lots of this broth. For these studies, the Autobac system and protocol have been used in conjunction with lyophilized preparations of the two reference organisms which have been sealed under vacuum in glass and stored at 4°C. Eugonic broth prepared in the Research lab from the same dehydrated lot using similar Production procedures serves as the time zero data points. The results of these studies are summarized in TABLE III and support a 16 month growth support stability for 4°C storage, and a 10 month growth support stability at room temperature. Storage at room temperature for ≥ 16 months leads to a decline in growth support particularly for E. coli ATCC 25922. 35°C storage for 9 months drastically reduces the growth support for both E. coli and S. aureus reference strains. Data collected on one lot which was prepared by sterile filtration and aseptic fill (lot 23009) as opposed to terminal autoclaving also shows 16 month stability at 4°C.

B. Turbidity Stability of the Final Product at 4°C and Room Temperature.

The 35° angle light scattering intensity, expressed as $-\log_{10}$ (scattering intensity at $\phi = 35^0$) in volts was measured on freshly produced lots of Eugonic Broth (33211-33214, and on Eugonic Broth stored up to 16 months at 4°C and room temperature. The range in this logarithmic light scattering parameter using the Autobac photometer and a 16 x 125 mm glass cell varies inversely with scattering intensity from approximately 4.0 for air to 3.0 for 0.45 micron filtered water to 3.0 for 0.45 micron filtered Eugonic Broth to ca. 2.0 for a 10^7 cell/ml. suspension of bacteria in 0.45 micron filtered Eugonic Broth. The stability data, shown in TABLE IV, indicates no measureable shift in the light scattering of Eugonic Broth on storage of up to 16 months at either 4°C or room temperature when compared with the typical range of scattering values obtained with freshly prepared broth lots.

TABLE IV

Production Prepared Eugonic Broth Lot No.	Time of Storage	Temperature of Storage	Autobac Light Scattering = $-\log_{10}$ (= 35° scattering intensity in volts
33211	ca 2 wks.	4°C	2.97 - 3.06^t
33212	"	"	2.95 - 2.98^t
33213	"	"	2.99 - 3.07^t
33214	"	"	2.97 - 3.10^t
22267	16 months	"	3.04*
22267	16 months	room temp.	2.93*
28155	10 months	4°C	3.00*
28155	10 months	room temp.	3.09*
23009	16 months	4°C	3.05*
23009	16 months	room temp.	3.02*
24060	10 months	room temp.	2.96*

t = range of 5 tubes; * = value for a single tube.

C. Integrity/Cleanliness of Screw Cap Liners After Storage.

Cap liners of tubes used in the stability studies of Sections I and II of this report concerning phosphate-buffered saline and Eugonic broth were examined for physical integrity and cleanliness after the longest storage period for any lot at any temperature. The cap liners of both products were found to be fully firm, functional, and to be clean regardless of the storage time or temperature. One exception to the cleanliness observation was noted for some tubes of lot 23C09 (produced by aseptic fill) which appeared to contain a small amount of hardened media on the liner.

J. E. McKie, Jr.

J. E. McKie, Jr.

JM/bv

[1]Also referred to as Inoculum Standardization Solution.

SECTION 6a

Details of analytical procedures for all active ingredients. The analytical procedures should be capable of determining the active components and of assuring the identity of such components.

All active ingredients (antibiotics) are purchased from reputable suppliers who have on file with us an appropriate Antibiotic Form 4. All are from FDA certified lots, with potency designated. These assays are accepted if confirmed by assay. Prior to first use, each lot is assayed vs. a control lot. This control may be a primary standard provided by FDA, or a primary or secondary (working) standard provided by a reputable supplier. The assay methodology requires that disks prepared from the test lot be compared to standard disks prepared from the standard lot. The comparison testing is carried out with the assay organism and assay procedure designated in the appropriate section of 21 CFR, 147.1.

A new lot of bulk antibiotic is acceptable only if it yields zones comparable to the control lot.

SECTION 4b

Standard used for acceptance of each lot of the finished drug.

Disk Assay Procedure: Assays are conducted by the standard procedure prescribed by FDA for that disk in 21 CFR, 147.1, or by alternate assays of established equivalency. Thus, for disks with an acceptable range of 68-150% of label or broader, the first day assay is within one Standard Deviation, approximately 15%, of the intended value, a second day assay may be omitted. (A single day assay has already been approved for standard diffusion disks.) For disks with acceptable ranges less than 68-150% of label, assays will be sufficiently replicated to yield results of adequate accuracy.

Standard Curve points are varied from the CFR recommendations in some cases, as shown in Table 5, attached to this section. Additional modifications, described in the attached report by F. C. Keenan, are used for Ampicillin 0.23 mcg. and Penicillin 0.2 U. In addition to meeting relevant requirements of CFR, 147.1, the specifications for "Elution Disks for the Autobac 1 System" are satisfied.

The nominal (label) potency, and the permissable range for which approval is requested are listed in Table 12. (Substantiating data for these potencies and ranges are presented in CFR 167.3, Section II. G of this application).

If the lot assays satisfactorily, samples are submitted on Antibiotic Form 7 to the National Center for Antibiotics and Insulin Analysis for assay and to the Division of Certification Services, FDA, for certification.

SPECIFICATIONS

RELEASE - R

6771

ORIGINAL

1 or 2

Carbon Disks for the Autobac 1

21 CFR 147.1.e.1

CFR 147.11

CONFIDENTIAL

SPECIFICATIONS

RELEASE - R

6/71 | ORIGINAL

2 of 2

Blot for Holes for the Anti-Sun

CER 547.2

CER 547.2

CONFIDENTIAL

A Microbiological Assay Procedure for the Assay of anti biotic Diffusion Plates for the Autobac I

Antimicrobial Agent	Nominal Potency	Optimal Potency Range, % of Nominal	Number of Curves	Number of Plates ea. Curve	Number of Disks ea. Plate
fe[illegible]illin	1.6 mcg.	80-125	3	5	6
ici[illegible]illin	0.32 mcg.	80-130	3	5	6
[illegible]tracin	18 U.	80-150	1	3	2
benicillin	120 mcg.	80-150	1	3	2
bolothin	15 mcg.	68-150	1	3	2
oramphenicol	4 mcg.	80-150	1	3	2
ndamycin	2 mcg.	68-150	1	3	2
[illegible]tin	11 mcg.*	65-150	1	3	2
cycline	1.6 mcg.	80-130	1	5	6
cycline	0.5 mcg.	68-130	1	3	2
bromycin	2.5 mcg.	68-150	1	3	2
anicin	0 mcg.	68-180	1	3	2
mycin	22 mcg.	80-150	1	3	2
omycin	2.4 mcg.	68-150	1	3	2
ici[illegible]illin	5 mcg.	68-180	1	3	2
dixic Acid*	15 mcg.	68-180	1	3	2
ycin	24 mcg.	68-150	1	3	2
ofurantoin*	15 mcg.	80-125	3	5	6
biocin	2.5 mcg.	68-180	1	3	2
nomycin	6 mcg.	68-150	1	3	2
cillin G	0.2 U.	68-130	1	3	2
ycin B	12.5 U.	68-150	1	3	2
ptomycin	30 mcg.	80-150	1	3	2

* assay is preferred.

A Microbiological Assay Procedure for the Assay of Antibiotic Potency Disks for the Autobac I

Antimicrobial Agent	Nominal Potency	Optimal Potency Range, % of Nominal	Number of Curves	Number of Plates ea. Curve	Number of Disks ea. Plate
Tetracycline	1.2 mcg.	80-130	3	5	6
Tetracycline	0.5 mcg.	68-150	1	3	2
Vancomycin	10 mcg.	68-180	1	3	2

Potency Standards and Coded Concentration Disks

Disk	Potency mcg(U)	Code		Standard Curve Points used, mcg(U)					Differs from CFR
Ampicillin (1)	0.22	AM	ep	.05	.10	.25	.5	1.0	Yes
Ampicillin (1)	1.6	AM	en	1.3	2.4	4.4	8.1	15.0	No
Bacitracin	18	B	ep	3.3	6.3	12.2	23.4	45	No
Carbenicillin	120	CB	en	33	63	122	234	450	Yes
Cephalothin	15	CL	ea	15	21.2	30.0	42.4	60.0	No
Chloramphenicol	4	C	en	3.3	6.3	12.2	23.4	45.0	No
Clindamycin	2	CM	ep	0.33	0.63	1.22	2.34	4.50	Yes
Colistin	13	CS	en	1.3	2.4	4.4	8.1	15.0	No
Doxycycline	0.5	DX	ep	0.33	0.63	1.22	2.34	4.50	Yes
Doxycycline	1.6	DX	en	"	"	"	"	"	"
Erythromycin	2.5	E	ep	1.30	2.70	5.40	11.0	22.5	No
Gentamicin	9	GM	ea	1.3	2.4	4.4	8.1	15.0	No
Kanamycin	22	K	en	3.3	6.3	12.2	23.4	45.0	No
Methicillin	5	SC	ep	1.3	2.4	4.4	8.1	15.0	No
Nalidixic Acid[a]	15	NA	en	3	6	12	24	48	N.R.(2)
Neomycin	24	N	en	3.3	6.3	12.2	23.4	45.0	No
Nitrofurantoin	15	FD	en	3	6	12	24	48	N.R.(2)
Novobiocin	2.5	NV	ep	1.00	1.41	2.00	2.82	4.00	Yes
Oleandomycin	6	OL	ep	1.3	2.7	5.4	11.0	22.5	No
Penicillin G (1)	0.20	P	ep	.05	10	.20	.50	1.0	Yes
Polymyxin B	12.5U	PB	en	10	12.5	15	20	25 (3)	Yes
Streptomycin	20	ST	en	3.3	6.3	12.2	23.4	45.0	Yes
Tetracycline	0.5	TE	ep	0.33	0.63	1.22	2.34	4.50	Yes
	1.2	TE	en	"	"	"	"	"	"
Vancomycin	10	VA	ep	3.3	6.3	12.2	23.4	45.0	No

(1) See separate report for details of assay

(2) N.R. - No CFR method exists. Our assay procedure for Nitrofurantoin and Nalidixic Acid resemble the CFR 147.1 method. Details are presented under "Ancillary Data."

(3) Tentative values, to be confirmed.

Autobac 1 Antibiotic Elution Disks and Permissable Potency Ranges for which approval is being sought in this application

Disk	Label Potency mcg., (U.)	Permissable Range % of Label
Ampicillin	3.6	80-125%
Carbenicillin	120	80-150
Cephalothin	15	68-180
Chloramphenicol	4	80-150
Clindamycin	2	68-150
Colistin	13	68-150
Erythromycin	2.5	68-150
Gentamicin	9	68-180
Kanamycin	22	80-150
Methicillin	5	68-180
Penicillin G	0.20.	68-180
Polymyxin B	12.50.	68-150.
Tetracycline (G+ organisms)	0.5	68-150
Tetracycline (G- organisms)	1.2	80-130
Vancomycin	10	68-180

BROOKLYN QUALITY CONTROL
Development Laboratories

January 31, 1973

TO : Dr. D. M. Cohen

FROM : F. C. Keenan

SUBJECT: ASSAY DEVELOPMENT FOR PENICILLIN G AND AMPICILLIN SENSITIVITY DISCS - OCSA 50765.

SUMMARY

Assays were developed for 0.25 mcg. Ampicillin and 0.2 unit Penicillin G sensitivity discs. The assays correspond in method with those used by the FDA and incorporate Staphylococcus aureus ATCC 6538 P as the test organism. Reproducibility of zone sizes, clarity of zones and suitable sensitivity and curve spread were obtained with assay ranges of .05 → 1.0 units of Penicillin G or micrograms of Ampicillin.

DISCUSSION

At the request of Pfizer Diagnostics, procedures were developed for the agar plate assay of 0.2 unit Penicillin G and 0.25 mcg. Ampicillin sensitivity discs. Within the framework of the CFR assay methods, trials were run utilizing both Sarcina lutea ATCC 9341, and Staphylococcus aureus ATCC 6538 P as test organisms. Both gave clear, well defined zones of inhibition when tested. Since less inoculum was required and better zone size spread between concentrations was obtained with the S. aureus strain, it was judged best for use in both the Penicillin G and Ampicillin assays.

Assay Procedure for Penicillin G and Ampicillin Sensitivity Discs

1. Culture Media:

a) Seed Agar (Medium A) BBL-10937
b) base Agar (Medium E) BBL-10943

The media used were dehydrated preparations whose ingredients are listed in the CFR section 147.1.

N 50580 -9

DATA

Zone sizes for replicate plates are presented in Tables I and II for Penicillin G and Ampicillin assays respectively. Representative curves using a straight line equation formula are depicted after the Tables (Graphs I and II).

CONCLUSIONS

Suitable assays have been developed for low level sensitivity discs of Ampicillin and Penicillin G. The use of S. aureus as the test organism for both assays gives clear, reproducible zones in the concentration ranges needed. As requested, the assays have been developed within the framework of the CFR. Detailed procedures have been presented so that the assays can be duplicated when needed.

F. C. Keenan

FCK:tc

CC: Dr. H. F. Hammer

Mr. F. J. Carleton
Ms. N. E. Dowd
Dr. J. Hackett ✓
Mr. W. B. Hardie
Dr. F. A. Hochstein
Mr. K. P. Hunnelly
Dr. J. Praglin

TABLE I

Penicillin G Disc Assay Results - Replicate Plates
Using S. aureus (Batch U02) 2.0 cc/L

Zone Sizes in mm.

Standard Curve Concentrations in Units/Disc.					Samples			
.05	0.1	0.2	0.5	1.0	(Theoretical Value - 0.2 Units/Disc			
13.5	18.5	22.0	27.5	30.0	22.0	23.0	21.5	22.0
14.0	18.5	21.5	26.5	30.0	22.5	22.0	21.5	22.0
14.5	17.5	27.0	26.0	29.5	22.5	23.0	21.5	22.0
14.5	17.5	22.0	26.5	29.0	23.0	23.0	23.0	22.5
15.0	18.0	22.5	27.0	29.0	23.0	22.5	23.0	22.0
$\bar{X}$ 14.3	18.0	22.0	26.7	29.5				

Disc potency = .24 Units/Disc.

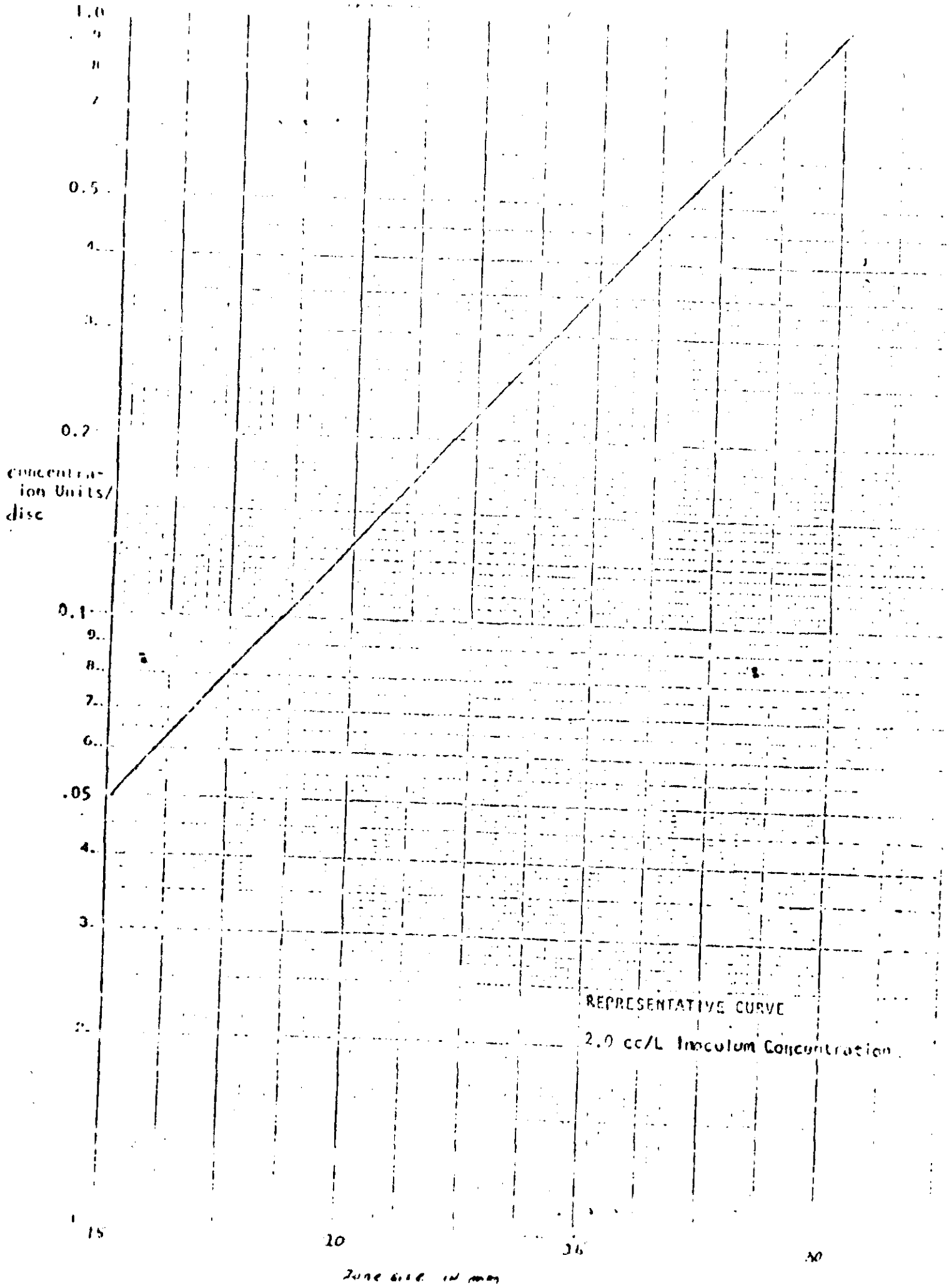
TABLE II

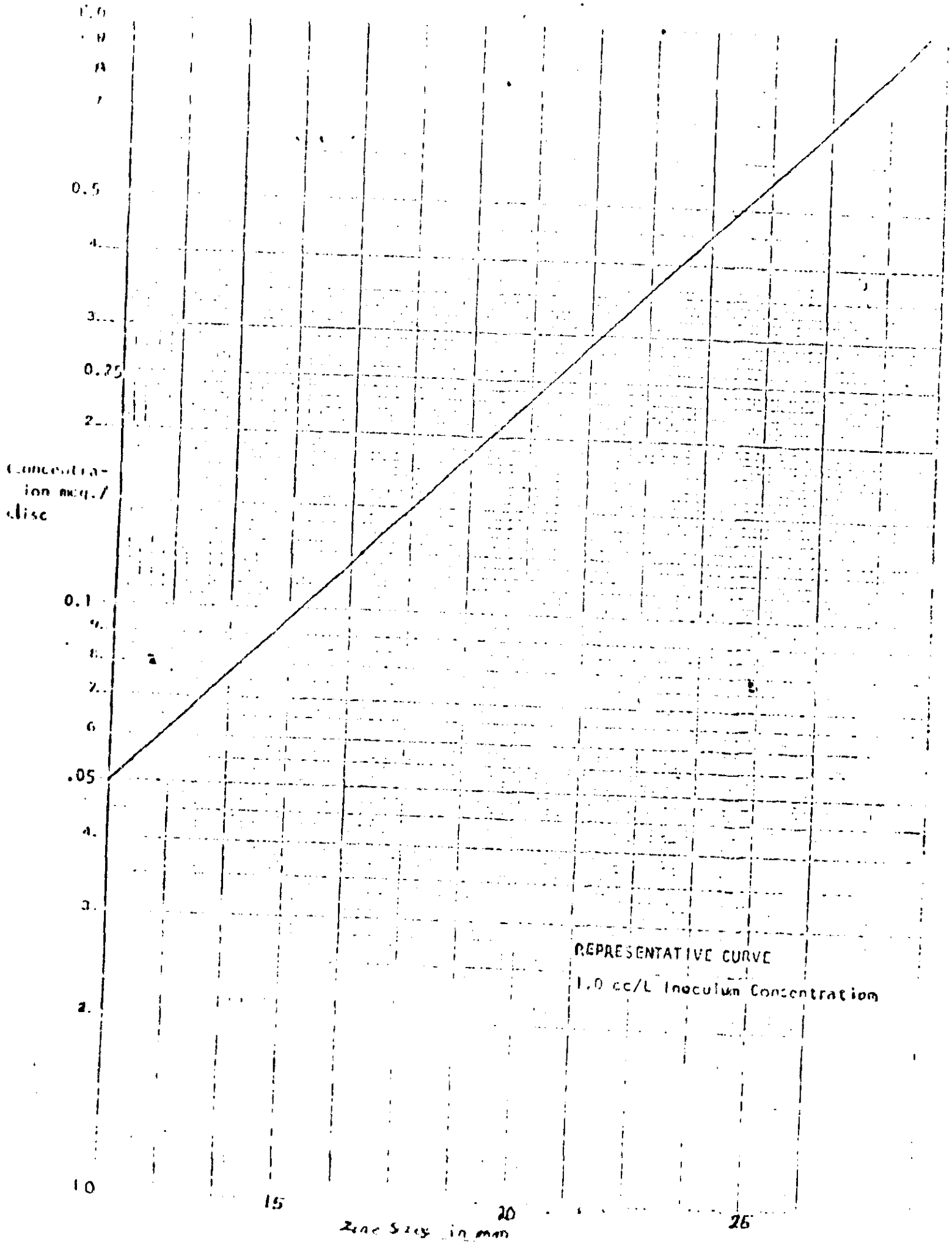
Ampicillin Disc Assay Results - Replicate Plates
Using S. aureus (Batch U02) 1.0 cc/L

Zone Sizes in mm.

Standard Curve Concentrations in mcg./Disc.					Samples			
.05	0.1	0.25	0.5	1.0	(Theoretical Value - 0.25 mcg./Disc.			
11.0	15.0	20.0	24.5	27.5	21.0	22.0	22.5	22.0
10.5	14.5	21.0	24.0	28.5	21.5	21.0	20.5	21.5
11.0	15.0	21.5	25.0	28.5	22.0	22.5	22.5	22.5
11.0	15.5	22.5	24.0	27.5	22.0	23.0	22.5	21.5
10.0	15.0	21.0	24.0	28.0	23.0	21.5	22.5	22.0
$\bar{X}$ 10.7	16.0	21.2	24.3	28.0				

Disc potency = .32 mcg./Disc.


1.0
0.5
0.2
concentration Units/disc
0.1
.05
15
20
25
30
REPRESENTATIVE CURVE
2.0 cc/L Inoculum Concentration


0.5
4
3
0.25
2
0.1
.05
2.
10
15
20
25
Zone Size in mm
REPRESENTATIVE CURVE
1.0 cc/L Inoculum Concentration

SECTION 4c

A detailed description of the collection of the samples to be tested by the applicant and by the Food and Drug Administration.

Following impregnation, strips are dried on "screens", which are loaded successively in known sequence. Following drying, strips are randomly selected from each screen. Disks are then punched from these selected strips in a manner which yields two to four disks from each strip per cartridge. Thus a vial of 100 disks represents 25-50 strips, which were in turn selected from 13-50 screens. All disks used for internal assay purposes, or for submission for certification are randomly selected from this group of cartridges.

This sampling procedure has been approved by FDA, and has been in use for several years.

SECTION 4d

Copies of all printed forms used by the applicant in the laboratory control of raw ingredients and the finished batch.

Copies of forms used are attached. See also Section 3a for additional forms.

REQUEST FOR ASSAY

Disks Date

...ne	Lot No.	Assay Type Required		
		FDA	CANADA	

Date Completed

by

Remarks:

[illegible] Date

NOTICE OF BATCH RELEASE

Lot Number

Name & Label Potency

Date Certified (Or Released)

FDA Certificate No.

FDA Potency

Pfizer Potency

Expiration Date Assigned

Assigned By

SENSITIVITY DISK ASSAY DATA SHEET

DRUG NAME:

Date Start _____ Date End _____ ASSAY BY: _____

Potency: Disks for Standard Curve	Standard Disk Zone Sizes - Millimeters: Plate 1	Plate 2	Plate 3	Plate 4	Plate 5		Avera Zone S
						A	
						B	
						C	
						D	
						E	

$$L = \frac{3A + 2B + C - E}{5} = \frac{3(\quad) + 2(\quad) + (\quad) - (\quad)}{5} =$$

$$H = \frac{3E + 2D + C - A}{5} = \frac{3(\quad) + 2(\quad) + (\quad) - (\quad)}{5} =$$

Plate No.	Test Disk Zone Sizes - Millimeters Lot No. _____ Label Potency _____		
1			
2			
3			
4			

TEST DISK ASSAY RESULTS

Total of Zone Sizes _____ mm

Average of Zone Sizes _____ mm

Uniformity (Largest less Smallest) _____ mm

Potency This Assay _____

Average Potency of Lot _____

Remarks: (Indicate if Sterility OK)

SECTION 4e

A complete description of the laboratory facilities used in such controls, including:

(i) The location of the laboratory in relation to the plant where the drug is manufactured,

(ii) A description of the laboratory equipment available for performing tests and assays, and

(iii) The names of the persons who will be responsible for conducting the required laboratory tests and information concerning their scientific training and experience.

The Quality Control Laboratory is located at Pfizer Diagnostics antimicrobial susceptibility disk manufacturing facility at 199 Maywood Avenue, Maywood, New Jersey.

The laboratory occupies upwards of 1400 square feet, and is fitted with standard laboratory benches and all facilities appropriate for disk assay. These include balances, pH meters, colorimeters, refrigerators, freezers, incubators, autoclaves, media preparation facilities. This facility was last inspected, in conjunction with our manufacture of antimicrobial susceptibility disks for diffusion assays, in February, 1973.

Additional laboratory facilities, which provide a wider range of general laboratory instrumentation, including e.g. ultraviolet and infra-red spectrophotometers, are located at Eastern Point Road in Groton, Connecticut. These laboratories are not used for the routine assay and control of certifiable disks. They may be utilized for the assay of non-certifiable disks.

SECTION 4c

A complete description of the laboratory facilities used in such controls, including:

(i) The location of the laboratory in relation to the plant where the drug is manufactured,

(ii) A description of the laboratory equipment available for performing tests and assays, and

(iii) The names of the persons who will be responsible for conducting the required laboratory tests and information concerning their scientific training and experience.

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The laboratory occupies upwards of 1400 square feet, and is fitted with standard laboratory benches and all facilities appropriate for disk assay. These include balances, pH meters, colorimeters, refrigerators, freezers, incubators, autoclaves, media preparation facilities. This facility was last inspected, in conjunction with our manufacture of antimicrobial susceptibility disks for diffusion assays, in February, 1973.

Additional laboratory facilities, which provide a wider range of general laboratory instrumentation, including e.g. ultraviolet and infra-red spectrophotometers, are located at Eastern Point Road in Groton, Connecticut. These laboratories are not used for the routine assay and control of certifiable disks. They may be utilized for the assay of non-certifiable disks.

RICHARD B. DARDAS, PH. D.
MANAGER MICROBIOLOGY QUALITY CONTROL
PFIZER DIAGNOSTICS
MAYWOOD, NEW JERSEY

EDUCATION:

B.A. Biology	1957	Albion College
M.S. Microbiology	1959	Michigan State University
Ph.D. Immunochemistry	1963	Michigan State University

EXPERIENCE:

Research Assistant Michigan State University	1957 - 1959
Research Fellow, Michigan State University	1959 - 1963
Staff Immunologist, Pfizer	1963 - 1967
Supervisor Chemical Res. & Dev., Pfizer	1967 - 1970
Supervisor Quality Control, Pfizer	1970 - 1972
Manager Quality Control Microbiology	1973 -

PUBLICATIONS:

Two in the field of microbiology

Joseph L. Hackett, Ph. D.

Supervisor, Microbiology Quality Control
Pfizer Diagnostics
Maywood, New Jersey

Education

B. Sc., Medical Technology	Ohio State University	1959
M. Sc., Clinical Pathology	Ohio State University	1963
Ph. D., Clinical Pathology	Ohio State University	1968

Experience

Research Assistant Infectious Diseases Laboratory Ohio State University Hospital	1960-1967
Quality Control Manager, Courtland Scientific Products Division, Abbott Laboratories	1967-1969
Microbiology Section Head, Reference Laboratories, North Hollywood, California	1969-1972
Supervisor, Microbiology Quality Control	1972-Present

Publications: Total of 4 in fields of infectious diseases

SECTION 41

If the applicant uses the services of a consulting laboratory, the name and address of such laboratory and a statement from such laboratory that includes the information required under 4(a), (b), and (c).

No consulting laboratories are used.

SECTION 4g

An explanation of the exact significance of any batch numbers used in the manufacturing, processing, packaging, and labeling of the drug, including such control numbers that may appear on the label of the finished article. State whether these numbers enable determination of the complete manufacturing history of the product. Describe any methods used to permit determination of the distribution of any batch if its recall is required.

A batch lot number is assigned to each batch of impregnated strips. This number is obtained from a master bound book which lists all disks made in chronological order. The numbers in this master book are in numerical sequence. The production sheets for that batch are marked with the designated batch lot number and the book is marked with the drug name, potency and date of manufacture. This batch lot number is likewise marked on the racks containing the strips which are being dried, the containers in which dried strips are stored, the sheets bearing the assay data for the batch, and the boxes in which packages of disks from this batch are stored. After the batch has been certified and released in writing by Quality Control, the assigned expiration date is entered into the master book. When packages of disks are labelled, the same lot number and expiration date are imprinted onto each label used and appropriate records maintained in the Label Accountability Book.

Each batch lot is fully described and identified on an Individual Production Sheet - Sensitivity Disks - and on Disk Disposition Sheets (see attached). The batch lot number will permit tracing the complete production history of the lot.

The lot number shipped in response to each order is recorded in a manner which permits tracing the complete distribution of each lot, in the event a recall is required.

See also Section 3q.

SECTION 4h

A complete description of, and data derived from, stability studies of the potency and physical characteristics of the drug, including information showing the suitability of the analytical methods used. Describe any additional stability studies underway or contemplated. Stability data should be submitted for any new antibiotic, for the finished dosage form of the drug in the container including a multiple-dose container in which it is to be marketed, and if it is to be put into solution at the time of dispensing, for the solution prepared as directed.

Stability Studies.

For fifteen of the certifiable elution disks intended for use in the Autobac I system, diffusion disks of substantially equivalent or lower nominal potency than the corresponding elution disk are currently manufactured for diffusion assays. For these fifteen disks, we claim expiration dates equal to those established for the diffusion disks. (Discussions with Dr. W. Wright, Messrs. G. Carter and D. Norton in November, 1972, led to informal approval of this proposal.) For the remaining certifiable disks, stability studies are underway on three lots.

Table II summarizes the potencies of disks now manufactured for diffusion assays, those proposed for use with the Autobac I system, the expiration dates of approved disks and the expiration dates claimed for disks manufactured identically except for adjustment to higher potencies. References are provided also to the Form 6 applications which contain data substantiating these claims. Stability data for the five disks of potencies significantly lower than are now certified and for which approval is requested by this application are presented in Table 13, following.

The assay methodology used is described in the introductory text. This methodology is identical to that described in 21 CFR 147.1, modified where necessary to allow accommodation of the higher or lower potencies involved. For all disks except Ampicillin 0.25 mcg. and Penicillin 0.2 U. the only change is in the potency of the standards to that designated in Table 5 of the introductory text. For the two disks listed above, additional changes were made, as described in a report included in Section 4b.

The presently available stability data on Cephalothin 15 mcg., Penicillin 0.2 U., Tetracycline 0.5 and 1.2 mcg., and Polymyxin B 12.5 U., for which approval is requested by this application, is shown in the attached Table 12. For all other disks for which approval is being sought at this time, we rely, as stated above, on stability data provided earlier. Those disks for which approval is being sought <u>now</u> are marked by marginal arrows in Table II.

Stability studies are underway on three lots of each disk for which new data is required. It will be reported periodically.

TABLE II

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form Numb
	Ampicillin	2		18		60-4
		10		18		
			.22		18	
--->			3.6*		18	
	Bacitracin	2U.		60		60-5
		10U.				
			18		60	
	Carbenicillin	50		24		61-4
--->			120*		24	
	Cephalothin	30		24		60-9
--->			15*		18	
	Chloramphenicol	5		24		60-3
		30				
--->			4*		24	
	Clindamycin	2		24		61-3
--->			2*		24	
	Colistin	2		24		60-9
		10				
--->			13*		24	
	Doxycycline	5		36		60-9
		30				
			.5		12	
			1.6		12	

SECTION 3 PAGE 3

TABLE II Con't.

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 1/74	Exp. Date Claimed for Elution Disks, mos.	Form Numb
	Erythromycin	2 15		24		60-9
--->			2.5*		24	
	Gentamicin	10		18		60-9
--->			9*		18	
	Kanamycin	5 30		24		60-9
--->			22*		24	
	Lincomycin	2	2.4	48	48	60-9
	Methicillin	5		18		60-9
--->			5*		18	
	Nalidixic Acid	5 30		N.A. (36)		-
-			15		36	
	Nitrofurantoin	100 300		N.A. (36) N.A. (36)		-
			15		to be estab.	
	Neomycin	5 30		30		60-99
			24		30	
	Novobiocin	5 30		36		60-99
			2.5		12	
	Oleandomycin	2 15		24		60-99

SECTION 8 PAGE 439

TABLE II (Con't.)

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form 6 Number
	Penicillin G	2U. 10U.		12		60-997
--->			0.2 U*		12	
	Polymyxin B	500. 300U.		36		60-999
--->			12.5 U*		to be estab.	
	Streptomycin	2 10		24		61-000
			20		24	
	Tetracycline	5 30		24		61-002
--->			0.5*		12	
--->			1.2*		18	
	Vancomycin	5 30		36		61-003
--->			10*		36	

---> * Approval is being sought for these disks by this application

TABLE 10

Stability Data on Antibiotic Susceptibility Disks for the Autoread I

Drug	Nominal Disk Conc. (mcg)	Lot No.	Assays at subsequent times - months								
			Original	1 mo.	2-3 mo.	4 mo.	5 mo.	6 mo.	7-8 mo.	9 mo.	Date (Lot)
Penicillin	10	2271	5/10/72 11					12/72 11		2/73 11	2/73 (2271) 11 11
		2433	12/15/72 19		2/73 19				2/73 19		2/73 (2433)19
		2524	12/15/72 15		2/73 19				2/73 19		2/73 (2524)19
		2532	3/73 22	3/73 16			5/73 19				5/73 (2532)19
Penicillin G	0.4U	2267	5/12/72 0.49							2/73 0.32	5/73 (2267)0.46
	0.2U	2388	10/4/72 0.20				3/73 0.23			2/73 0.29	12/73 (2388)0.30
	0.2U	2385	3/9/72 0.25		6/73 0.18	7/73 0.27					
	0.2U	2451	2/8/72 0.28			6/73 0.26	7/73 0.30				12/73 (2451)0.32
	0.2U	2467	2/8/72 0.28			6/73 0.28					12/73 (2467)0.30
Tetracycline	0.5	2373	10/4/72 0.60				3/73 0.61	4/73 0.59			12/73 (2373)0.61
	0.5	2462	3/17/73 0.65	4/73 0.69							12/73 (2462)0.68
	0.5	2468	3/17/73 0.60	4/73 0.68							12/73 (2468)0.69
	1.0	2278	6/17/72 1.4							3/73 1.2	4/73 12/73 (2278)1.2 (2278)1.1
	1.0	2437	3/17/73 1.2	4/73 1.2							12/73 (2437)1.3
	1.0	2499	3/17/73 1.3	4/73 1.2							12/73 (2499)1.0
Polymyxin B	12.5U	2285	7/12/72 12				12/72 13				

SECTION 41

The expiration date needed to preserve the identity, strength, quality and purity of the drug until it is used.

For disks equal to or greater in potency than currently certified diffusion disks, we claim expiration dates allowed for diffusion disks as indicated in Table 11, following, and substantiated by data on file with the indicated Form 6. For disks of lower potency, we claim an expiration date of six months, based on the information presented in Table 12, Section 6h.

All disks for which an expiration date is being sought in this application are marked by a marginal arrow in Table 11.

TABLE II

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	For Num
	Ampicillin	2 10		18 18		60-
--->			.22 3.6*		18 18	
	Bacitracin	2U. 10U.		60		60-
			18		60	
	Carbenicillin	50		24		61-
--->			120*		24	
	Cephalothin	30		24		60-
--->			15*		18	
	Chloramphenicol	5 30		24		60-
--->			4*		24	
	Clindamycin	2		24		61-
--->			2*		24	
	Colistin	2 10		24		60-
--->			13*		24	
	Doxycycline	5 30		36		60-9
			.5 1.6		12 12	

TABLE II (Con't.)

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form Numb
	Erythromycin	2 15		24		60-9
--->			2.5*		24	
	Gentamicin	10		18		60-9
--->			9*		18	
	Kanamycin	5 30		24		60-9
--->			22*		24	
	Lincomycin	2	2.4	48	48	60-9
	Methicillin	5		18		60-9
--->			5*		18	
	Nalidixic Acid	5 30		N.A. (36) N.A. (36)		-
			15		36	
	Nitrofurantoin	100 300		N.A. (36) N.A. (36)		-
			15		to be estab.	
	Neomycin	5 30		30		60-99
			24		30	
	Novobiocin	5 30		36		60-99
			2.5		12	
	Oleandomycin	2 15		24		60-99

TABLE II (Con't.)

Comparison of Diffusion and Elution Disk Potencies and Claimed Expiration Dates

	Antimicrobial Agent	Nominal Potency Diffusion Disks, mcg.(U.)	Nominal Potency Autobac 1 Elution Disks, mcg.(U.)	Exp. Dating Allowed for Diffusion Disks, mos., 2/74	Exp. Date Claimed for Elution Disks, mos.	Form 6 Number
	Penicillin G	2U. 10U.		12		60-997
--->			0.2 U*		12	
	Polymyxin B	50U. 300U.		36		60-999
--->			12.5 U*		to be estab.	
	Streptomycin	2 10		24		61-000
			20		24	
	Tetracycline	5 30		24		61-002
--->			0.5*		12	
--->			1.2*		18	
	Vancomycin	3 30		36		61-003
--->			10*		36	

---> * Approval is being sought for these disks by this application

SECTION 5a,b,c,d,e

The following samples shall be submitted with the application or as soon thereafter as they become available.

a) If it is a new antibiotic: 10 grams of the applicant's reference standard if an official standard has not been designated, plus 5 grams from each of three separate batches. Include for any reference standard a complete description of its preparation and the results of all laboratory tests on it. If the test methods differed from those described in the application, full details of the methods employed in obtaining the reported results shall be submitted.

b) If it is a dosage form: 6 immediate containers (or 30 tablets or capsules) from each of three separate batches, except that if it is a sterile drug 30 containers shall be submitted from each of three batches.

c) Include for samples submitted pursuant to items 5(a) or 5(b) detailed results of all laboratory tests made to determine the identity, strength, quality and purity of the batch represented by the sample.

d) Additional samples shall be submitted on request.

e) The requirements of items 5(a) or 5(b) may be waived in whole or in part on request of the applicant, or otherwise, when any such samples are not necessary.

Sample Submissions:

By prior agreement we have already submitted three lots of each of the following certifiable disks for which approval is requested in this application. This group includes those disks whose nominal assay value is significantly higher or lower than presently certified disks, and for which minor modification of the assay procedure in CFR 147.1 might be required.

Identity	Nominal Potency	% Optimum Potency Range	Lot No.	Submission Date	Pfizer Assay	FDA Assay
Carbenicillin	120 mcg.	80-150	2291	6/21/73	117	132
			2531	"	142	129
			2577	"	132	131
Cephalothin	15 mcg.	68-180	2271	12/8/72	17.7	16.
			2423	3/27/73	18.9	20.
			2424	"	18.1	22.
Colistin	13 mcg.	68-150	2252	12/8/72	12.7	12.
			2463	3/27/73	15.3	13.
			2469	"	13.6	14.
Penicillin	0.2 U.	68-180	2385	3/27/73	0.25	0.2
			2461	"	0.28	0.3
			2467	"	0.28	0.3
Polymyxin	12.5 U.	68-150	2285	12/8/72	11.6	
			2482	7/17/72	17.1	
			2483	"	16.0	
Tetracycline	0.5 mcg.	68-150	2373	5/31/73	0.61	0.61
			2462	"	0.69	0.71
			2468	"	0.68	0.79
	1.2 mcg.	80-130	2278	5/31/73	1.15	1.21
			2497	"	1.16	1.40
			2499	"	1.20	1.82

*Difficulty was encountered in assaying this disk due to irregular zones. New disks, providing substantial circular zones, are in preparation.

Copies of letters forwarded with the disks already submitted are appended.

We will provide samples of all disks not already submitted, and of new Polymixin and Tetracycline disks on request.

DIAGNOSTICS DIVISION
100 MAYWOOD AVENUE MAYWOOD N J 07607

December 8, 1972

Mr. Gordon G. Carter
Chief, Antibiotic Residue Branch
National Center for Antibiotics Analysis
Bureau of Drugs, HD-437
200 C Street SW
Washington, D. C. 20204

Dear Mr. Carter,

On November 2nd. Dr. F. A. Hochstein and other representatives from the Pfizer Diagnostics Research Group discussed with you our automated device for determining antibiotic susceptibility. You may recall that the device uses antibiotic susceptibility disks which differ in potency from those now certified for the Kirby-Bauer diffusion assay. You may also recall that you asked to receive disks in advance of any formal submission to facilitate the development or adoption of the diffusion assays.

We are, therefore, sending you under separate cover the following disks at this time.

Identity	Code	Lot No.	Nom Potency	Assay 6-72	Assay 11-72
Cephalothin	CLe	2271	15 mcg	17.7 mcg	20.9 mcg
Colistin	CSe	2252	13 mcg	12.7 mcg	9.98 mcg
Polymyxin-B	PBe	2285	12.5 mcg	11.6 mcg	

All assays were run by the standard CFR assay. We have started to repeat assays for the assessment of stability . We will, if you wish, provide you with assay values as they are performed.

Sincerely yours,

J. L. Hackett, Ph. D.
Microbiology
Quality Control

JLH:db

RECEIVED
DEC

DIAGNOSTICS DIVISION
188 MAYWOOD AVENUE, MAYWOOD, N.J. 07607

March 27, 1973

Mr. Gordon G. Carter
Chief, Antibiotic Residue Branch
National Center for Antibiotics Analysis
Bureau of Drugs, BD-437
200 C Street SW
Washington, D. C. 20204

Dear Mr. Carter:

We are sending additional susceptibility disks for use in the Pfizer automated device for measuring antibiotic susceptibility. The disks are provided to enable you, at your convenience, to develop assays for these disks, some of which differ in potency from the disks currently certified for the Kirby-Bauer diffusion assay. These disks are provided in advance of formal submission.

Included in the samples are low potency Ampicillin and low potency Penicillin disks. A suggested method for the assay is enclosed.

We are aware of irregularities in the zones of inhibition produced with the low potency Polymyxin disks. We are currently working on developing an assay to correct the abnormalities.

Under separate cover we are sending you the following:

Identity	Code	Lot No.	Nom. Potency	Assays
Cephalothin	CLe	2423	15 mcg.	18.9 mcg. [2/73]
Cephalothin	CLe	2424	15 mcg.	18.1 mcg. [2/73]
Colistin	CSe	2463	13 mcg.	15.3 mcg. [1/73]
Colistin	CSe	2469	13 mcg.	13.6 mcg. [1/73]
Penicillin	Pe	2385	0.2 u.	0.25 u. [3/73]
Penicillin	Pe	2461	0.2 u.	0.26 u. [2/73]
Penicillin	Pe	2467	0.2 u.	0.28 u. [2/73]
Ampicillin	AMe	2354	0.25 mcg.	0.24 mcg. [9/72]
				0.18 mcg. [3/73]
Ampicillin	AMe	2459	0.25 mcg.	0.29 mcg. [2/73]
Ampicillin	AMe	2465	0.25 mcg.	0.27 mcg. [2/73]

Sincerely yours,

J. L. Hackett, Ph. D.
Microbiology
Quality Control

JLH:db
Enc.

DIAGNOSTICS DIVISION
100 MAYWOOD AVENUE MAYWOOD N.J. 07607

RECEIVED
JUN 5 1973
F. A. HOCHSTEIN

May 31, 1973

Mr. Gordon G. Carter
Chief, Antibiotic Residue Branch
National Center for Antibiotics Analysis
Bureau of Drugs, BD-437
200 C Street SW
Washington, D. C. 20204

Dear Mr. Carter:

We are sending you two (2) additional antiblotic susceptibility disks for use in the Pfizer automated device. The disks are provided in advance of formal submissions to enable you to develop assays. Both of these antiblotics - Tetracycline and Doxycycline - are of lower potency than those currently certified.

In preparing the standards we used the enclosed.

Under separate cover we are sending you the following:

Identity	Code	Lot No.	Non Potency	Assays
Tetracycline	TEe	2373	0.5 mcg.	0.61 mcg. [4/73]
Tetracycline	TEe	2462	0.5 mcg.	0.69 mcg. [4/73]
Tetracycline	TEe	2468	0.5 mcg.	0.68 mcg. [4/73]
Tetracycline	TEe	2278	1.0 mcg.	1.15 mcg. [4/73]
Tetracycline	TEe	2497	1.0 mcg.	1.16 mcg. [4/73]
Tetracycline	TEe	2499	1.0 mcg.	1.20 mcg. [4/73]
Doxycycline	DXe	2384	0.5 mcg.	0.59 mcg. [5/73]
Doxycycline	DXe	2460	0.5 mcg.	0.65 mcg. [5/73]
Doxycycline	DXe	2466	0.5 mcg.	0.67 mcg. [5/73]
Doxycycline	DXe	2293	2.0 mcg.	2.03 mcg. [5/73]
Doxycycline	DXe	2496	2.0 mcg.	2.72 mcg. [5/73]
Doxycycline	DXe	2500	2.0 mcg.	2.64 mcg. [5/73]

It should be pointed out that at the moment, we feel the optimal potency for the Tetracycline disks shoul[d] [1].5 mcg. and 1.5 mcg., and for the Doxycycline disks 0.5 mcg. and 1.6 mcg. [Al]though the higher potency disks do not match these concentrations we feel they are well within the range to develop an adequate assay.

Sincerely yours,

J. L. Hackett, Ph. D.
Microbiology
Quality Control

JLH: db
Enc.

TETRACYCLINE - DOXYCYCLINE

Stock Solution prepared so that 0.02 ml contained 4.5 mcg.

Stock Solution 0.02 ml = 4.5 mcg.

20 ml = 4,500 mcg.

Amount of drug weighed out = $\frac{4{,}500}{\text{Activity of Standard}}$

= Grams hydrated in 20 ml solvent (methanol)

Standard Disk Potency	Dilutions of Stock Solution	Solution No.	Amt. of Solvent to be added to 1 ml of Solution #5
0.33 mcg.	1:13.64	1	12.64 ml
0.63 mcg.	1:7.15	2	6.15 ml
1.22 mcg.	1:3.69	3	2.69 ml
2.34 mcg.	1:1.92	4	0.92 ml
4.50 mcg.	1:1	5	—

Zone sizes of the different reference point standards are approximately as follows:

	Tetracycline	Doxycycline
0.33 mcg. Std disk	11-12 mm	12-13 mm
0.63 mcg. Std disk	14-15 mm	14-15 mm
1.22 mcg. Std disk	16-17 mm	16-17 mm
2.34 mcg. Std disk	19-20 mm	19-20 mm
4.50 mcg. Std disk	22-23 mm	22-23 mm

DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
WASHINGTON, D.C. 20204

June 1, 1973

RECEIVED
JUN 4 1973
F. A. HOCHSTEIN

J. [illegible] Beckett, Ph.D.
Microbiology Quality Control
Pfizer Diagnostics Division
[illegible] Maywood Avenue
Maywood, New Jersey 07607

Dear Dr. Beckett:

In response to your letters of December 7, 1972, and March 27, 1973, with the subsequent submission of [illegible]s of antibiotic "elution" discs, we have performed a number of assays of these materials and have evaluated several disc agar-diffusion methods.

This letter is intended to outline the present status of the elution disc assay methods and to provide you with our analytical results as of this date.

Attached are the assay results of 12 batches of elution discs representing 4 drugs. Cephalothin and colistin assays were performed with the present CDC methods which have been modified, in each case, by using standard doses of 3.75, 7.5, [illegible] and 30 µg per disc. Penicillin and ampicillin assays were performed using the [illegible] agar diffusion method provided by your laboratories. [illegible] of 0.05, 0.1, 0.2, 0.4, and 0.8 µg per disc were used for ampicillin and 0.05, 0.1, 0.2, 0.4, and 0.8 units per disc for penicillin. These doses were selected so that they would be equally spaced logarithmically.

We experienced no particular assay problems with these batches of discs. However, you are aware of the assay difficulties we experienced with the one lot of low level polymyxin discs. I understand your laboratories are working to improve this assay procedure.

Dr. Hochstein informed me today that we would be receiving some doxycycline and tetracycline elution discs with which we may check the tetracycline assay method. He mentioned that the drug concentrations of these discs would not equal that level which may ultimately be proposed, but that the drug level would be in the appropriate range for elution discs. Dr. Hochstein also stated that we soon should be receiving a revised polymyxin assay procedure which has eliminated most, but not all, of the irregular shaped zones previously experienced with this assay.

If you have any questions about the assay methods or if I can provide you with any additional information, please do not hesitate to call.

Sincerely yours,

Gordon G. Carter, Chief
Antibiotic Residue Branch
National Center for Antibiotic
Analysis, HD-437
200 'C' Street, S. W.
Washington, D. C. 20204

cc: Dr. W. W. Wright, BD-400
Dr. P. J. Weiss, BD-430
Mr. R. Norton, BD-140
Reading File
Dr. F. Hockstein, Pfizer Inc.

[illegible]

Label = 15 µg

	Lot 2459	Lot 2465	Lot 2252
Day 1	15.1	1[illegible].0	12.2
	16.0	15.0	13.2
Day 2	15.3	12.5	11.5
	14.9	12.5	11.5
Day 3	14.7	13.5	
	14.6	13.6	
Mean	14.9 µg	13.5 µg	12.1 µg

CEPHALOTHIN DISCS

Label = 15 µg

	Lot 2425	Lot 2424	Lot 2271
Day 1	19.4	21.1	16.0
	21.2	22.4	16.5
Day 2	20.0	19.5	[illegible]
	21.8	24.0	17.0
Day 3	[illegible].6	24.4	
	22.2	21.6	
Mean	20.9 µg	22.2 µg	16.5 µg

AMPICILLIN DISCS

Label = 0.25 µg

	Lot 2361	Lot 2462	Lot 2459
Day 1	0.25	0.38	0.35
	0.27	0.33	0.35
Day 2	0.26	0.33	0.34
	0.26	0.33	0.35
Day 3	0.28	0.36	0.35
	0.29	0.35	0.35
Mean	0.27 µg	0.36 µg	0.34 µg

PENICILLIN DISCS

Label - 0.20 unit

	Lot 2385	Lot 2461	Lot 2367
Day 1	0.25	0.30	0.33
	0.25	0.31	0.31
Day 2	0.28	0.33	0.33
	0.28	0.[illegible]	0.32
Day 3	0.26	0.34	0.30
	0.26	0.29	0.3[illegible]
Mean	0.26 u.	0.32 u.	0.32 u.

DIAGNOSTICS DIVISION

June 21, 1974

Mr. Gordon C. Carter
Chief, Antibiotic Residue Branch
National Center for Antibiotics Analysis
Bureau of Drugs, HEW
200 C Street SW
Washington, D. C. 20204

Dear Mr. Carter:

We are sending you an additional antibiotic susceptibility disk designed for use in the Pfizer automated sensitivity testing device. The disks of Carbenicillin have a nominal potency of 120 mcg.

In the assay of the disks we use the following standards: 33, 63, 122, 238 and 450 mcg.

Zone sizes vary from approximately 15 mm for the 33 mcg. standard to approximately 22 mm for the 450 mcg. standard.

Under separate cover we are sending you the following:

Identity	Code	Lot No.	Nom. Potency	Assay
Carbenicillin	CBe	2691	120 mcg.	117 mcg. (4/4)
Carbenicillin	CBe	2531	120 mcg.	142 mcg. (3/3)
Carbenicillin	CBe	2577	120 mcg.	132 mcg. (4/4)

You will also find enclosed a report from our development group regarding the assay of the Polymyxin B disks. The report lists several modifications of the FDA assay which enables the diffusion assay to be more precise. In preparing the inoculum the following is recommended.

After growing the organism in the usual manner the organism is washed off the slant and adjusted so that a 1:20 dilution of the bulk in saline will give 31% transmission at 530 mμ. The dilution is placed in a 16 x 100 mm tube and inserted into the well of a model 401 Lumetron. The undiluted bulk is added to the seed layer in a volume of 0.5 - 1.0 ml/liter of agar.

Sincerely yours,

J. L. Wachett, M. D.
Microbiology
Quality Control

DIAGNOSTICS DIVISION

July 17, 1973

Mr. Gordon G. Carter
Chief, Antibiotic Residue Branch
Regional Center for Antibiotics Analysis
Bureau of Drugs, BD-137
200 C Street SW
Washington, D. C. 20204

Dear Mr. Carter:

We are sending you two additional lots of Polymyxin B. The disks are intended for use in the Difco automated sensitivity testing device. The two lots, being sent to you under separate cover, are as follows:

Identity	Code	Lot No.	Nom. Potency	Assay
Polymyxin B	Pbe	2180	12.5 u.	17.1 u. (6/73)
Polymyxin B	Pbe	2185	12.5 u.	16.0 u. (6/73)

I refer you to my letter of June 21, 1973 in regard to the modified FDA assay procedure.

Sincerely yours,

J. L. Hackett, Ph. D.
Microbiology
Quality Control

DEPARTMENT OF HEALTH, EDUCATION AND WELFARE
FOOD AND DRUG ADMINISTRATION
WASHINGTON, D.C. 20204

July 31, 1973

J. L. Hackett, Ph.D.
Microbiology Quality Control
Pfizer Diagnostics Division
199 Maywood Avenue
Maywood, New Jersey 07607

RECEIVED
AUG 6 1973
F. A. HOCHSTEIN

Dear Dr. Hackett:

In response to your letters of May 31, June 21, and July 17, 1973, with subsequent submission of 17 batches of elution discs, we have performed agar-diffusion assays of these materials and have evaluated Pfizer's polymyxin disc assay method dated June 6, 1973.

Attached are the assay results of the 15 batches of discs representing 3 drugs. The tetracycline and doxycycline assays were performed with the present CFR S. lutea method which was modified, in each case, by using standard doses of 0.33, 0.63, 1.22, 2.34, and 4.50 µg per disc. For purposes of evaluating the S. lutea agar diffusion method we assayed the doxycycline elution discs even though this drug is not included in the present regulations for susceptibility discs. The carbenicillin assays were performed with the present CFR Pseudomonas aeruginosa method which was modified by using standard doses of 10, 20, 40, 80, 160 µg per disc.

A review of the June 6th agar-diffusion assay method for 12.5 unit polymyxin discs indicates you are proposing preincubation for 3 hours at 5°C, incubation at 34°C instead of 37°C, the preparation of standard doses in 10% pH 6 phosphate buffer instead of distilled water. Also you specify that the discs should not be treated with any material that either enhances or inhibits the activity of the polymyxin, your data show that the response lines of standard discs made from these two solvents are indeed significantly different. We have observed this same difference in response lines in our laboratory. We know that variations in salt concentration of buffers greatly alters the diffusion of polymyxin in the cylinder-plate assay. We used your proposed method and still observed a large number of irregularly-shaped zones of inhibition. I must conclude that there is presently no satisfactory agar-diffusion assay method for polymyxin elution discs. I would be interested in receiving your comments concerning this matter.

Our laboratory will retain the two batches of polymyxin elution discs (lots 2482 and 2483) for future assays. The first lot (2285) of polymyxin elution discs submitted in December 1972, was discarded after the initial analytical problems. We will need a portion of another production lot to complete the testing when the polymyxin assays problems are solved.

If you have any questions regarding the analytical results, please do not hesitate to call.

Attachments

Sincerely yours,

Gordon G. Carter
Chief, Antibiotic Residue Branch
National Center for Antibiotic
Analysis, BD-437
200 'C' Street, S. W.
Washington, D. C. 20204

cc:
Dr. Frank Hockstein, Pfizer Inc.

TETRACYCLINE ELUTION DISCS
Label = 0.5 mcg

	lot 2373	lot 2462	lot 2468
Day 1	0.63	0.66	0.68
	0.63	0.69	0.69
Day 2	0.59	0.78	0.88
	0.61	0.74	0.89*
Day 3	0.58	0.70	0.72
	0.62	0.68	0.72
Day 4			0.82
			0.84
			0.76
			0.91*
Mean	0.61	0.71	0.79
% label	122	142	158

* total of 2 discs out of 60 were non-uniform.

CARBENICILLIN PLATTEN DISCS
Label = 120 µg

	lot 2301	lot 2531	lot 2577
Day 1	130	125	130
	130	130	142
Day 2	135	124	126
	135	136	126
Mean	132	129	131
Label	110	108	109

DOXYCYCLINE ELUTION DISCS
Label = 0.5 mg

		lot 2384	lot 2460	lot 2466
Day 1		0.62	0.66	0.75
		0.63	0.64	0.72
Day 2		0.70	0.74	0.80
		0.76	0.74	0.84
	Mean	0.68	0.70	0.78
	% Label	136	140	156

Label = 2.0 mg

		lot 2393	lot 2496	lot 2500
Day 1		2.10	3.21	3.04
		2.06	3.15	3.20
Day 2		2.04	3.04	3.22
		2.04	3.04	3.22
	Mean	2.06	3.11	3.17
	% Label	103	156	158

TETRACYCLINE ELUTION DISCS
Label = 1.0 mcg

	lot 2278	lot 2497	lot 2499
Day 1	1.34	1.45	1.98*
	1.34	1.45	1.82*
Day 2	1.10	1.35	1.69
	1.05	1.33	1.99
Day 3			1.95*
			1.61
Day 4			1.74
			1.77
Mean	1.21	1.40	1.82
% label	121	140	182

* total of 4 discs out of 48 were non-uniform.

9. HAZARDS TO USER

9. HAZARDS TO USER

A. Potential Microbiological Hazards

B. Potential Hazards and Safeguards Relative To Hardware

POTENTIAL MICROBIOLOGICAL HAZARDS

The potential health hazard associated with the improper handling of pathogenic microorganisms in any microbiological methodology should be recognized. In this respect, microorganisms subjected to Autobac I testing should be manipulated by trained personnel using the same accepted handling methods as used in any microbiological methodology.

Since the Autobac I method has a number of procedures, as well as hardware unique to this susceptibility testing method, potential hazards associated with each step of the method will now be outlined.

Step 1 Standardization -

a) In the preparation of the standard suspension of bacterial cells in the phosphate-buffered saline, care should be taken to avoid the deposition of bacteria from the loop onto the interior of the threaded neck of the standardization tube. If this does occur, one should be careful not to touch these bacteria with the pipette in the broth inoculation step.

b) After the bacteria have been loop transferred to the buffered saline, the standardization tube should be tightly capped (with the threaded closure provided) before vortexing. Vortexing the tube with a loose cap can lead to spattering. After reading the tube in the photometer, should dilution be required DO NOT attempt to add buffered saline to the tube while it is in the photometer.

Step 2 Introduction of Antimicrobial Disks -

a) Although the desired panel of elution disks may be dropped into the broth inoculum filled cuvette, we recommend loading the cuvette with the disk panel prior to introducing the broth inoculum. This is advised for reasons of both convenience (cuvette cap has to be seated only once in the latter method and the empty cuvette does not have to be maintained in a reasonably level orientation during handling) as well as safety (although quite remote, there is a possibility that the cuvette cap could become contaminated by improperly distributing the broth inoculum into the cuvette; removal of this cap to accomplish disk addition could lead to contamination of the hands, desk top, etc)

b) After dispensing the elution disk panel into the empty cuvette, place the cuvette cap securely on the cuvette. Briefly check each chamber to insure that the corresponding nipple of the cap is seated into the chamber opening. In practice, if the broth inoculum is properly distributed (see Step 3) in a cuvette without a cuvette cap, no broth inoculum will spill out of the chamber openings. The presence of a cap primarily serves as a safeguard against spillage if the cuvette is not handled properly or is accidentally dropped.

Step 3 Broth Inoculum Preparation and Distribution -

a) When pipetting the aliquot of buffered saline inoculum into the broth tube a cotton-plugged pipette should be used. It is recommended for additional safety that a pipetting bulb be used, rather than pipetting by mouth.

b) The buffered saline tube with its remaining inoculum and the tube cap should be discarded into an appropriate pathogen container immediately after use. Similarly, the pipette should be placed immediately after use into an appropriate pathogen container.

c) The inoculated broth tube should be capped tightly, gently mixed by several inversions, uncapped and after discarding the broth tube cap into a pathogen container, the tube screwed into the cuvette port until it is snugly fitted against the sealing washer in the port.

d) The broth inoculum is then distributed to the thirteen cuvette chambers by three rotational manipulations of the cuvette on a level surface. Although the cuvette is specifically designed so that no broth inoculum will leak out during these rotations, there is always a finite chance that a leak will be encountered. A supply of 5 vol. % phenol or 70 vol. % ethanol should be readily available so that the contaminated working area can be flooded in such an event. A soft tissue soaked in either of the ethanol or phenol solutions can be used to clean the cuvette if only a minor leak has occurred (i.e., $\leq$ 0.5 ml.). If a larger leak is encountered, the cuvette and contents should be discarded into an appropriate pathogen container for incineration or autoclaving (see Step 6).

Step 4 Incubation/Agitation -

a) Cuvettes should be securely positioned onto the brackets located on each tray.

b) If any leakage from a cuvette occurs during the incubation/agitation period, the individual trays and interior of the incubator can be decontaminated by washing down with 5 vol. % phenol, 70 vol. % ethanol, or other appropriate antiseptics.

Step 5 Reading -

If for any reason bacterial contamination occurs in the reading chamber of the photometer, it is designed to be easily decontaminated by wiping down with 5 vol. % phenol, 70 vol. % ethanol, or other appropriate antiseptic.

Step 6 Disposal of Cuvettes -

After completion of susceptibility readings, the broth-filled cuvette with its attached broth tube are disposed of by autoclaving at 121°C for 30 minutes or by incineration.

While in the above discussion all potential hazards and their avoidance is stressed, it should also be recognized that the Autobac I methodology requires that for most of the procedure, pathogen-containing tubes and cuvettes are tightly sealed.

In this respect, chance contamination of the laboratory using the Autobac I methodology is less likely than using current manual techniques.

POTENTIAL [illegible]S
AND SAFEGUA[illegible]IVE
TO U[illegible]

The Autobac system is powered by a three wire grounded line cord and protected by a circuit breaker switch. The ground wire is firmly attached to the chassis and all metal parts are electrically connected to the same chassis.

The system is designed such that tools are required to remove covers to gain access to any electrical wiring.

All power supplies exposed for service purposes are isolated from the utilities by a double shielded transformer.

The instrument is designed so that in the course of normal operation, all dangerous electrical connectors are covered with aluminum plates or plastic covers to insure safety. If the covers must be removed in order to service the instrument, it is necessary to switch off electrical power and to disconnect the instrument from the utility outlet to insure safety during repair operations.

The electrical components located under the blue top cover behind the printer operate at a temperature of 220° F. If it becomes necessary to remove this cover, caution must be exercised to prevent contact with these components.

The quartz halogen lamp behind the cuvette carriage operates at a temperature of 600° F., and the lamp housing at a temperature of 210° F. If the lamp requires servicing, avoid contact with the lamp and the lamp housing, and handle the lamp only by its leads, since fingerprints will damage the bulb and cause it to burn out.

N 50580 Bio -1

AD-A150580

DTIC

aztreonam IV
[redacted]TAM injection
NDA 50-5[redacted]
1 g powd[redacted]al for
reconstitution
Reviewer: I. Gonzalez

60580

E.R.Squibb & Sons, Inc.
Route 1 at College Farm Rd.
Submission date of
Supplement # S-006: Feb. 23, 1987
Supplement # S-009: May 18, 1987

[redacted]

Review of Amendments to NDA

Background:
Aztreonam is a synthetic bactericidal monobactam antibiotic with activity against a wide spectrum of gram negative aerobic pathogens. Aztreonam for injection is a sterile, nonpyrogenic white powder. AZACTAM for injection contains 1 gram of aztreonam with approximately 780 mg L-arginine, for reconstitution before IV administration.

Aztreonam is indicated for the treatment of urinary tract infections, lower respiratory tract infections, septicemia, skin and skin structure infections, intra-abdominal and gynecologic infections.

The supplements which are the subject of this review have been submitted in support of the use of aztreonam for the treatment of meningitis caused by susceptible gram-negative organisms, for the use of aztreonam in pediatric populations and in cystic fibrosis patients.

Studies in support of meningitis (adults):

A. Protocol #18554-29:

Title: Single IV Dose Safety and Pharmacokinetic Study of Aztreonam in Patients with Normal or Inflamed Meninges.

Investigator/Site: Richard Duma, M.D./Medical College of Virginia.

Supplies: Aztreonam 1000 mg powder for injection Lot # MNB-864-H/B31, B33 and B36 was supplied by the sponsor.

Design:
A 5-min IV infusion of a single 2000 mg dose was given to 30 (25 evaluable) adult patients (21-75 yrs) with normal meninges (Group A) and to 10 (9 evaluable) patients (19-71 yrs) with gram-negative bacterial or viral meningitis (Group B). Exclusion criteria was found appropriate. Patients in group B, received aztreonam in addition to the antibiotic program prescribed by their physician.

Specimens collected:
A 15 ml bl[redacted] sample was collected at: prior to dose, 30 min after end of infusion and at time of spinal tap. A 4 ml CSF sample was collected at: within 7 days prior to dose and at time of spinal tap.

Assay methodology:
Serum and CSF samples were assayed by HPLC and by a microbiological assay method. Assays have been previously reviewed and were found acceptable. A good correlation was found between the results obtained by the different methods. Plot of concentrations obtained by bioassay versus those obtained by HPLC had slope=1. Results were based on the bio-assay data.

Sponsor results:
Table 1, page 5-011, contains mean serum and CSF concentrations at time of spinal tap. Figures 5 and 6 depict concentration-time plots for both treatment groups (Table and Figures are reproduced in Appendix I to this review).

Mean CSF levels of aztreonam in the absence of meningitis were 0.5 and 1 ug/ml at 1 and 4 h, respectively, and, were 2 and 3.2 ug/ml at 1 and 4 h, respectively, in the presence of meningitis. Meningeal inflammation produced approximately 3 to 4 times higher CSF levels of aztreonam than values in the absence of inflammation.

Serum data in patients with normal meninges (0.5 to 8 h) was described by a monoexponential equation. The elimination half-life in serum was approximately 1.7 h. CSF data in the same patients was described by a biexponential equation. Due to the limited data available for patients with meningitis, pharmacokinetic analysis for this group was not possible. The CSF penetration (ratio of AUCs) of aztreonam between 0.5 and 8 h after the 2 g dose averaged 1.5% in the absence of meningeal inflammation.

Conclusions:
In adult patients with meningitis, CSF levels were higher and serum levels lower compared to those in patients with normal meninges. Peak CSF levels occurred 2 to 4 h after drug administration. CSF levels were at least 1 ug/ml (except in one case) between 2 and 8 h.

A 2000 mg dose in adults produced CSF concentrations that are potentially therapeutic for members of "enterobacteriaceae" commonly responsible for gram-negative bacillary meningitis.

B. Study Protocol 18554-51:

Title: Single intravenous Dose Safety and Pharmacokinetic Study of Aztreonam in Patients with Inflamed Meninges.

Investigator/Site: The study was conducted by Richard Greenman M.D. at University of Miami School of Medicine, and Leon L. Sabath, M.D. at University of Minnesota School of Medicine.

Supplies: Aztreonam 1000 mg powder for injection Lot #MNB-854-H/C01 was supplied by Squibb.

Design: This study was designed to complement Protocol 18554-29. The same dose (2000 mg IV over 5 min) was given to 11 additional patients with meningeal inflammation. Exclusion criteria and specimen sampling was as the previous study (18554-29).

Sponsor results:
Serum concentrations of aztreonam averaged 101, 41 and 15 ug/ml at 0.5, 2 and 4 h after dose, respectively, in this group of patients (meningitis). Aztreonam was detectable in CSF at 0.5 hr after dose. The mean CSF concentrations were 1.36, 2.79, 4.60 and 3.31 ug/ml at 1, 2, 4 and 8 h after dose administration (See Table 1 in the study report, reproduced in Appendix I, attached to this review). Maximal CSF concentrations occurred between 2-4 h after dosing.

Conclusions:
Studies #18554-29 and 18554-51 were acceptably conducted. These studies have demonstrated that aztreonam was present in CSF of adult (21-75 yr) patients with meningitis at concentrations above 1.0 ug/ml during the interval between 1 and 8 h after the administration of 2000 mg (approx. 30 mg/kg) IV infusion over 5 min. Aztreonam concentrations in CSF of adult patients with meningitis were 3 to 4 times higher than in adult (19-72 yr) patients with normal meninges.

In the presence of meningitis, CSF penetration of aztreonam (ratio of AUC(CSF)/AUC(Serum)) will, most likely, be greater than 1.5% (the value obtained in adult patients without meningitis) since CSF aztreonam levels were higher and serum levels were lower in adult (21-75) patients with meningitis compared to those with normal meninges.

Comment:
1. On May 3, 1988, through a telephone conversation between Mr. John Hunt and Dr. Larry T. Friedhoff, the firm was requested to provide plots of CSF levels in a graph showing the MIC_{90} of relevant pathogens. (See discussion of responses in section I, below.)

Studies in support of pediatric use:

C. Protocol #18554-32:

Title: Single Intravenous Dose (30 mg/kg) Safety and Pharmacokinetic Study of Aztreonam (SQ 26776) in Pediatric Patients.

Investigator/Site: Melvin I. Marks, M.D./Oklahoma Children's Memorial Hospital, Oklahoma City, Oklahoma.

Supplies: Sterile vials containing powder blend of aztreonam (1000 mg) and L-arginine (780 mg) for constitution. Lot # MNB-864-H/C01.

Design:
Thirty one (29 evaluable) pediatric patients participated, including some with meningitis who had spinal taps performed for diagnostic reasons. They ranged in ages from 2 days to 11.4 years and weight from 0.84 to 38.4 kg. They were classified by age into 5 groups:

I. Newborn	(a)	< 1 week old and < 2500 g weight
	(b)	< 1 week old and > 2500 g weight
	(c)	1 week to 1 month old
II. Infants		> 1 month old to 2 yr old
III. Children		> 2 yr old to 12 yr old

Exclusion criteria were: a) history of allergy (penicillin, cephalosporins); b) abnormal hepatic or renal function; c) abnormal deviations from clinical laboratory values; d) presence of clinical findings or history of potentially disqualifying conditions, as per investigators judgement.

A single dose (30 mg/kg IV infused over 3 min) open study. Patients remained hospitalized for at least the 3 days of the study.

Blood (1-2 ml) was collected at: pre-dose, 15 min, 1, 3 and 6 h after the end of the 3 min infusion. Cumulative urine samples were collected at intervals: pre-dose, 0-3 h, 3-6 h, 6-12 h and 12-24 h after the end of the infusion. CSF (0.4 ml) was obtained at moment of spinal tap. All samples were assayed by The Squibb Institute.

Assay methodology: HPLC and microbiological assays were employed. Validation data and analysis for each assay are presented on pages 5-708 to 5-784 and were found to be acceptable. The assays have been previously reviewed.

Pharmacokinetic/Statistical Analysis:
AUTOAN and NONLIN were used for fitting mono- and bi-exponential equations to the C,t data. AUCs were calculated using the trapezoidal method and were extrapolated to infinity. Evaluation

of statistical differences in half-life, MRT, V_d and Cl_s for the 5 age groups was performed by ANOVA and Duncan's Multiple Range Test. Half-life and MRT were log transformed prior to analysis.

Sponsor results/conclusions:
Tables and figures summarizing the sponsor's results are reproduced in Appendix II, attached to this review.
The elimination of aztreonam was slowest in newborns <1 wk and < 2500 g (mean Cl_s 0.94±0.14 ml/min/kg, mean $t_{1/2}$=5.71±1.63 h) and most rapid in children 2 to 12 yrs (mean Cl_s=2.50±0.15 ml/min/kg, and mean $t_{1/2}$= 1.67±0.21 h). The effect of age on aztreonam elimination and serum clearance was observed in three distinguishable patterns: Newborns <1 wk and <2500 g (slowest elimination), newborns <1 wk and >2500 g to 1 month of age and infants 1 mo to <2 yrs (faster rate), and children 2-12 yr old (fastest elimination). These results may be explained by the differences in renal function (based on inulin clearance expressed in ml/min/sq.m, as shown on Table 27) of these three age groups. This is in accordance with normal development of renal function during later gestation and the early years of life.

For the initial investigation in pediatric patients with serious life-threatening infections requiring administration of the maximal dose, the following treatment regimens were suggested by the sponsor (see also Table 30, Appendix II):

	Dose to be Given at 6-Hr Intervals	Interval to be Used for a 30 mg/kg Dose
Newborns <1 wk & <2.5 kg	20 mg/kg	12
<1 wk & >2.5 kg 1 wk - 1 mo Infants >1 mo - 2 yr	30 mg/kg	6
Children >2 yr - 12 yr	50 mg/kg	4

For a constant dose, values for the dosing interval were obtained based on the following equation:

$$I_{peds\ age\ grp} = I_{normal\ adult} \times Cl_{s,normal\ adult}/Cl_{s,peds\ age\ grp}$$

where: $Cl_{s,normal\ adult}$=1.5 ml/min/kg (from published literature). For a fixed interval, the dose for the particular pediatric age group can be estimated by:

$$D_{peds\ age\ grp} = D_{normal\ adult} \times Cl_{s,peds\ age\ grp}/Cl_{s,normal\ adult}$$

The computation was based on data in adult patients with life threatening gram-negative infections, for whom clinical experience has shown that 2 gm IV q 6 h (about 30 mg/kg) is a reasonable treatment regimen. Lower daily doses (1 g q 8 or 12 h) than those in the table (lower dose or longer dosing interval) are recommended for less severely ill patients. The previous calculations assume that aztreonam follows linear kinetics in pediatric patients as it does in adults.

Mean serum levels of aztreonam would be expected to exceed the MIC_{90} values for all bacteria shown (Figure 1, in Appendix II, attached to this review) except for enterobacter cloacae and pseudomona aeruginosa, for approximately 8-12 hr in patients on all age groups, and those of enterobacter cloacae and pseudomona aeruginosa for about 4, 8 or 12 h depending on the age group.

Mean urinary concentrations of aztreonam were in the range of 2.1 to 20.8 ug/ml during 0.75 to 4.33 h after dose. These levels would exceed the MIC_{90} for most enterobacteriaceae.

Penetration of the drug into CSF of pediatric patients with meningitis (n=4) was comparable to that found in adults with inflamed meninges. CSF levels after the 30 mg/kg dose were between 2 to 20 ug/ml in 4 patients: newborn 1 wk-1 mo (n=2) and infant >1mo-2 yr (n=2) (Table 24).

Conclusion:

The pharmacokinetics of aztreonam were evaluated in 29 pediatric patients (5 age groups) after a single, 30 mg/kg dose was given as a 3-minute IV infusion. No other doses have been studied. Estimates of pharmacokinetic parameters were obtained.

The sponsor has proposed recommended dosing regimens for the 5 different pediatric age groups. Calculations were based on the average adult serum clearance (1.5 ml/min/kg), the normal adult dose (30 mg/kg), and the clearance estimated for each pediatric group. These dosing regimens have been incorporated into the revised proposed package insert (included in Appendix XI, p. 9 of 11).

Comments:

2. Note that according to the sponsor's calculations, the recommended dose for children 2-12 yrs with life-threatening infections should be 50 mg/kg every 6 h (200 mg/kg/day), to compensate for their ability to eliminate aztreonam more rapidly.

Because this dose is much higher than the 30 mg/kg X 4=120 mg/kg regimen which had been extensively studied in adults, the sponsor has conducted a study in pediatric patients 2-12 yrs to evaluate the safety and kinetics of aztreonam upon administration of a 50 mg/kg dose (See Study 18554-32 Addendum A).

3. A plot of CSF levels in a graph showing the MIC_{90} of relevant pathogens was requested to the firm by this Division (see review below).

4. Blood was sampled only up to 6 hr post-dose. Based on the apparent differences in half-life between the pediatric age groups, and given that blood concentrations should be followed for at least 4-5 times the half-life of the drug, blood samples should have been collected for at least 16-20 hrs after dosing, particularly in the younger (newborns) age groups, so as to permit adequate estimation of the elimination parameters in these pediatric groups. Therefore, the half-life values in the younger groups could be underestimated and the clearances could be overestimated.

5. The <u>in-vitro</u> bilirubin displacement study included in Appendix D of the report should be reviewed by a biochemist.

<u>D. Protocol 18554-32 Addendum A</u>:

<u>Title</u>: Intravenous Dose Safety and Pharmacokinetic Study of Aztreonam in Pediatric Patients Given a Single 50 mg/kg Dose. (Investigator/Site as in Protocol 18554-32)

This study was designed to complement Protocol 18554-32 and studied 6 additional pediatric patients (children 3-11.9 yrs) with normal renal function. Each patient received a single 3-minute infusion of aztreonam at a dose of 50 mg/kg. Tables and figures summarizing the results of the study are reproduced in Appendix II.

<u>Sponsor Results</u>:
Mean serum concentrations of aztreonam were 214, 109, 38.6 and 12.8 ug/ml at 0.25, 1, 3 and 6 h after dosing, respectively (n=5). Mean urine concentrations of aztreonam were 3297, 1660, 358 and 35 ug/ml during the 0-3, 3-6, 6-12 and 12-24 h collection periods, respectively. Mean pharmacokinetic parameters were:

	$t_{\frac{1}{2}}$ hr	MRT hr	$V_{d_{ss}}$ liters/kg	$V_{d_{ss}}$ liters/m^2	Dose (mg/kg)	Cl_s ml/min/kg	Cl_s ml/min/m^2	Study
MEAN	1.99	2.08	0.24	6.23	50	1.94	50.6	32 A
S.E.M.	0.23	0.21	0.04	0.60		0.24	1.0	
Mean (SEM)	1.67 (0.21)	1.93 (0.33)	0.29 (0.07)		30	2.50 (0.15)		32

The mean serum clearance, volume of distribution, terminal half-life and MRT did not differ significantly from the same parameters measured in the earlier study (18554-32) of 2-12 yr old patients given 30 mg/kg doses (Table 20, Appendix II). When expressed on a ml/min/sq.m basis, the serum clearance, found in children in this study also does not differ meaningfully from 50 ml/min/sq.m, the value observed in adults given mean doses of 42 mg/kg. These results imply that the pharmacokinetics of aztreonam in children (2-12 yr) are linear, as they are for adults. They are also consistent with the fact renal function for children above the age of 2 years, expressed as creatinine clearance per body surface area, does not differ from adult values.

Conclusions:

Pharmacokinetic parameters of aztreonam were estimated in children 2-12 years of age (n=6) after the IV administration of a 50 mg/kg dose as a single, 3-minute infusion. Values for Cl_s, $t_{1/2}$, $V_{d_{ss}}$ and MRT were not much different from those obtained after a single, 30 mg/kg, 3-min IV infusion to 5 patients in the same age group (Study 18554-32).

Study #18554-32-A has been acceptably conducted.

Studies #18554-32 and -32-A support the dosage recommendations for children 2-12 years proposed in the revised Package Insert (Appendix XI).

Comments:

6. The data obtained in this particular age group (children 2-12 yrs) tend to indicate that the kinetics are linear. This will be discussed further with the review of Study #18554-58-A (multiple dose study).

7. The results of this study tend to suggest that a dose of 50 mg/kg of aztreonam is necessary in this pediatric group (2-12 yrs age) to treat life-threatening infections due to their increased clearance compared to other pediatric groups studied before. However, the safety of this dose in a multiple dose regimen should be evaluated by the Medical Officer.

E. Protocol 18554-52 and Addendum A:

Title: Single Intravenous-Dose Pharmacokinetic and Safety Study of Aztreonam (SQ 26776) in Pediatric Patients Given 30 or 50 mg/kg.

Investigator/Site: Allan M. Arbeter, M.D., and Stanley Plotkin, M.D./Children's Hospital, Philadelphia, PA.

Supplies: Aztreonam (1000 mg) was supplied in sterile vials containing L-arginine (780 mg) blend, Lot #MNB-864-H/C01.

Design: Twenty-four (20 evaluable) pediatric patients, hospitalized for various infectious, congenital or perinatal disorders, participated. Exclusion and inclusion criteria were as in Protocol 18554-32 and were found to be acceptable.

Each patient received a single 30 minute infusion of azcreonam at a dose of 30 (n=8) or 50 (n=16) mg/kg. Age groupings were as in Protocol 18554-32. Blood (1-2 ml) was collected pre-dose, at end of 30 min infusion and at 1, 3 and 6 hours. Urine was collected at intervals: prior to dose, ~0.5-3, 3-6, 6-12 and 12-24 h post-dose. One specimen of CSF was collected at time of spinal tap in meningitis patients.

Assay methodology:
Serum, CSF and urine samples were quantitated by HPLC. The serum inhibitory power was determined by a microbiological assay. Both assays have been previously reviewed. Validation data and analysis for each assay were included on pages 5-708 to 5-784 and are found to be acceptable.

Pharmacokinetic/Statistical Analysis:
Serum concentration data were analyzed using moment analysis. Terminal elimination rate constants were derived from fitting one or two exponential equations to C,t data using AUTOAN and NONLIN with $1/Y_i$ weights. AUCs were obtained by the trapezoidal method. Evaluation of statistical differences in $t_{1/2}$, MRT, $V_{d,ss}$ and Cl_s for the 5 age groups was performed by ANOVA and Duncan's Multiple Range Test. Half-life and MRT were not normally distributed and were transformed to natural logarithm prior to analysis.

Sponsor Results:
Mean pharmacokinetic parameters and other results are reproduced in Appendix III, Tables 20-25.

In patients <1 week and >2500 g or in those >1 week to 2 years, irrespective of age, a 30 mg/kg dose produced mean serum concentrations of 82 and 15 ug/ml, 0.5 and 6.5 h after the beginning of the infusion, respectively. Similar patients, given doses 50 mg/kg, had 211 and 27 ug/ml concentrations at 0.5 and 6.5 h, respectively. Children aged 2-12 years who received 50 mg/kg had mean serum concentrations of 186 and 7.4 ug/ml at 0.5 and 6.5 h after the start of the infusion, respectively. Mean urine aztreonam concentrations exceeded 600 ug/ml in all patient groups for times up to 3.5 to 6.5 h after dosing.

Patients in groups <1 week and >2500 g or 1 week to 2 years, irrespective of weight, showed no dose dependency in pharmacokinetic parameters and, therefore, the results for 30 and 50 mg/kg doses were combined in these groups. Renal clearance values are only approximate because it was difficult to assure complete urine collections in these pediatric patients. Patients aged 2-12 yrs (given 50 mg/kg doses) had a greater serum clearance than

the younger patients. The increased serum clearance in the older patients was associated with both, increased renal and non-renal (Cl_R-CL_{NR}) clearance and a shorter serum terminal elimination half-life. The table that follows summarizes mean pharmacokinetic parameters derived from this study.

CATEGORY	DOSE (mg/kg)		$T_{\frac{1}{2}}$ (hr)	MRT (hr)	Cl (ml/min/kg)	Cl (ml/min/m²)	V (l/kg)
Age <1 Wk and Wt >2500 gm or Age 1 Wk - 2 Yrs	30	MEAN	3.37	4.50	1.53	27.5	0.37
		SEM	0.54	0.73	0.15	4.0	0.05
		N	6	6	6	6	6
	50	MEAN	2.97	3.97	1.71	32.6	0.30
		SEM	0.86	1.13	0.37	9.1	0.05
		N	6	6	6	6	6
	Combined 30 or 50	MEAN	3.17	4.23	1.62	30.0	0.34
		SEM	0.49	0.91	0.19	4.8	0.03
		N	12	12	12	12	12
Age 2-12 Yrs	50	MEAN	1.15	1.93	2.51	65.6	0.27
		SEM	0.17	0.09	0.29	2.2	0.04
		N	5	5	5	5	5

The mean concentrations observed in patients <1 week and >2500 g or aged 1 week to 2 years who received 30 mg/kg doses, were similar to the concentrations previously reported for such patients. The mean serum concentrations significantly exceeded the reported MIC_{90} for most Enterobacteriaceae (<1 ug/ml) and Pseudomonas (12 ug/ml) throughout the interval studied (0.5 to 6.5 h after initiation of the infusion). Similar patients given 50 mg/kg had proportionately higher serum concentrations.

For patients aged 2-12 years who received 50 mg/kg doses, the mean serum concentrations also exceeded the MIC_{90} for most Enterobacteriaceae throughout the interval studied. The MIC_{90} for Pseudomonas was exceeded at 3.5 hr after initiation of the infusion but not at 6.5 hrs. For patients receiving every-6-hour therapy with aztreonam, a repeat infusion would be administered 6 hours after the beginning of the preceding infusion and, therefore, the trough level expected during the multiple dose therapy would be slightly higher than the 6.5 hr value obtained after a the single dose in this study.

The mean urinary concentrations of aztreonam found in all patient groups exceeded the MIC_{90} for common gram-negative organism during all the time periods evaluated (including the 12-24 hr period).

The CSF concentrations observed in this study were quite similar to those observed in earlier s' ies of pediatric patients. Although the levels observed were variable, they all exceeded 1

ug/ml. (CSF levels were between 1.7 and 21.0 ug/ml during the interval between 1.17 and 3.5 hrs. In study 18554-32, the range of CSF levels was 2.3-20.8 during the period between 0.8 to 4.3 hr post dose).

Conclusions:
Studies #18554-52 and -52-A were acceptably conducted.

The current single dose studies give evidence to support the constancy of aztreonam pharmacokinetic parameters ($t_{1/2}$, Cl_s, MRT and V_{dss}) at two dose levels, 30 and 50 mg/kg, in pediatric patients <1 week (>2.5 kg) to 2 years and 2 to 12 years old. The studies: (a) give support to the linearity in the pharmacokinetics of aztreonam in the pediatric groups studied, and (b) support the dosing recommendations made by the sponsor in the revised Package Insert (Appendix XI).

Comments:
8. No conclusions can be drawn regarding patients <1 week who weighed less than 2500 g because of lack of suitable patients. The sponsor has not been able to confirm the pharmacokinetic results obtained in study #18554-32 in this age group.

9. It appears that a dose of 50 mg/kg is a reasonable dose to use in pediatric patients with infections caused by several Enterobactereaceae and Pseudomonas (mean serum levels exceeded the MIC_{90} of each respective species throughout an interval of approximately 6 hours after initiation of the infusion). However, the safety of aztreonam after the administration of 50 mg/kg in a multiple dose regimen needs to be evaluated (see comment 7 and General comments).

F. Protocol #18554-58 Addendum A

Title: Study of Aztreonam Pharmacokinetics in Pediatric Patients with Gram-Negative Infections (multiple dose study)

Investigator/Site: Melvin Marks, M.D., and Harris Stutman, M.D./ Oklahoma Children's Memorial Hospital, Oklahoma City, Oklahoma.

Design: Six pediatric patients with various systemic infections were enrolled. Aztreonam, 30 mg/kg was administered as 30-minute infusions every 6 or 8 hours. Four patients were aged 11-12 years and 2 patients were 0.5 to 0.67 years. Serum concentrations were measured at the beginning (Days 1 or 2 of therapy) and at the end (Days 5, 7 or 9 of therapy) of the study.

Sponsor's Results:
Appendix IV of this review includes Tables 4, 5 and 6, which summarize the results of the study. Mean pharmacokinetic parameters are included below:

AGE		C (μg/ml) BEG. (Day 1 or 2)	C (μg/ml) END (Day 5, 7 or 9)	C (μg/ml) BEG.	C (μg/ml) END	AUC (hr.μg/ml) BEG.	AUC (hr.μg/ml) END	Cl (ml/min) BEG.	Cl (ml/min) END	Cl (ml/min/kg) BEG.	Cl (ml/min/kg) END	$t_{\frac{1}{2}\beta}$ (hrs) BEG.	$t_{\frac{1}{2}\beta}$ (hrs) END
11-12	MEAN	108.6	106.6	8.1	4.8	292.5	262.4	79.1	87.4	1.8	1.9	1.6	1.6
yrs	SD	12.9	10.2	5.3	1.0	82.1	32.1	15.5	22.5	0.4	0.2	0.4	0.5
	N	4	4	4	4	4	4	4	4	4	4	4	4
0.5-0.67	MEAN	61.7	59.3	1.9	5.5	173.5	244.0	23.8	17.9	2.8	2.1	1.3	1.9
yrs	N	2	2	2	2	2	2	2	2	2	2	2	2

Conclusion:
Serum aztreonam concentrations at the beginning (day 1 or 2) and the end of the study (day 5, 7 or 9) were similar at each time point. Pharmacokinetic parameters calculated on Days 1 or 2 of the study were approximately the same as those calculated on Days 5, 7 or 9 of the study. This limited data indicate that the pharmacokinetics of aztreonam are not altered with repeated dosing in pediatric patients and significant accumulation does not occur when a 30 mg/kg IV infusion (over 30 min) is given every 6 or 8 hours to children 11-12 yr or to infants 0.5 to 0.67 yrs.

Comment:
10. Given the limited data (only 6 patients were studied), the conclusion seems appropriate (General comment #1 for a comparison of observed and projected steady state levels).

G. Protocol #18554-58 Addendum D

Title: Study of Aztreonam Pharmacokinetics in Premature Pediatric Patients with Suspected Gram-Negative Infections. (multiple dose study)

Investigator/Site: George H. McCracken, Jr., M.D./ University of Texas Health Science Center, Dallas, TX.

Supplies: Aztreonam Lot # MNB-864-H/C105, was provided in sterile powder/arginine blend containing 500 mg aztreonam per vial for reconstitution.

Design: Twenty six hospitalized premature infants, ranging in age from 1 to 4 days and in weight from 0.7 to 2.0 kg at the time of enrollment, were classified by birth weight into three groups: 500-1000 g, 1001-1500 g and 1501-2000 grams The infants received 30 mg/kg aztreonam administered as a 15-minute infusion every 12 hours along with ampicillin administered according to the regimen

prescribed by the patient's primary physician. Approximately 0.2 to 0.3 ml of blood was collected for assay 10 min before, at the end of the 15 min infusion and at 0.5, 1, 2, 4 and 8 hr after the end of the infusion. After the last dose, blood was also collected 10 min before the last infusion, at the end of the 15 min infusion and at 0.5, 2 and 8 hr after the end of the last infusion. The first spontaneously voided urine specimen after the end of the first aztreonam infusion was collected. No CSF was obtained.

Assay method: Serum and urine samples were analyzed by the clinical investigator using a microbiologic method. Difco Laboratories Antibiotic medium No. 1 was used and the pH was adjusted to 7.9. The test organism was E. Coli SC 12155.

Sponsor's Results: Appendix V contains tables summarizing the study results. See also deficiency and comment.

The serum aztreonam clearance was higher on Days 3 to 4 (1.06 ml/min/kg) than on Day 1 (0.75 ml/min/kg). The more rapid clearance on Days 3 to 4 was reflected in the serum elimination half-life which tended to decrease between the beginning (Day 1 $t_{1/2}$=8.4 hrs) and the end (Day 3-4 $t_{1/2}$=6.3 hrs) of the study. These changes in the pharmacokinetic parameters can probably be explained by the physiologic changes that accompanied maturation (of premature infants) during the period of the study.

The concentrations of aztreonam in the urine of patients enrolled in this study always exceeded the MIC_{90} of organisms considered sensitive to the drug.

Deficiency:
1. The sponsor has not provided sufficient details about the assay methodology. It is inferred that the bioassay used (for which only a general description was provided) was different than the one previously used by the sponsor and found acceptable by the agency. Validation data is necessary but none have been provided with the study report.

Conclusions:
Study #18554-58-D is unacceptable due to lack of appropriate assay validation data and results. However, the study design seemed appropriate and the conduction of the study appeared acceptable.

In this multiple dose study, mean plasma levels achieved on Day 1 (after the first 30 mg/kg dose of a q12h regimen, infused over 15 min) were comparable to those obtained on Study #18554-32 after a single 30 mg/kg dose infused over 3 min. This study also tends to indicate that, in Study #18554-32, the serum clearance was overestimated and the half-life was underestimated in this pediatric group.

Comment:
11. According to the revised (proposed) Package Insert, the sponsor does not intend to recommend the use of aztreonam in pediatric patients less than one month old. Therefore, at the present time, this study is not relevant to the proposed new indications. If in the future, the sponsor intends to use Study #18554-58-D in support of the use of aztreonam in premature newborns, then, the sponsor will be required to submit assay validation methodology and data corresponding to this study.

H. Protocol #18554-62 (Cystic fibrosis)

Title: Single Intravenous-Dose Safety and Pharmacokinetic Study of Aztreonam (SQ 26-776) in Patients with Cystic Fibrosis.

Investigator/Site: Stephen C. Aronof, M.D./ Rainbow Babies and Children's Hospital, Cleveland, Ohio.

Supplies: Aztreonam (1000 mg) vials, Lot # MNB-864-H/C01, containing a sterile powder blend of aztreonam and L-arginine for reconstitution.

Design: Enrolled were eleven patients, males (although female patients could have been included), between 10 and 18 yrs of age (mean=14 yrs), 22-47 kg weight (mean=35 kg) with cystic fibrosis. Exclusion criteria included: a) history of allergy to penicillin or cephalosporin, b) lactating or pregnant females, c) abnormal hepatic or renal function, d) exposure to beta-lactams or probenecid within 24 h prior to dosing, e) medically significant deviations from normal clinical laboratory values, f) history or presence of any clinical finding that in the opinion of the investigator could disqualify the patient from the study.

It was an open study in which each patient received a single 30 mg/kg (not exceeding 2 g) infused IV over 3 min.

A 2-ml blood sample was drawn at the following times: prior to dose, 5, 15 and 30 min, 1, 2, 4, 6 and 8 h after the end of the 3-min infusion. Cumulative urine collections were obtained at: prior to dose, 1-2 h, 2-4 h, 4-8 h, 8-12 h, 12-24 h after the drug infusion.

Assay Methodology:
Serum and urine samples were assayed by the Squibb Institute. Aztreonam and its open ring metabolite, SQ 26-992, were assayed by HPLC (reviewed previously and found acceptable). Samples were also assayed for aztreonam by the clinical investigator using a different method.

Pharmacokinetic/Statistical Analysis:
Moment analysis was used to analyze the serum concentration-time

data. AUTOAN and NONLIN were used to fit mono- and bi-exponential equations to the C,t data using $1/Y_i$'s as weights. AUCs were calculated by the trapezoidal method and extrapolated to infinity. Half-lives, clearances, volume of distribution at steady state and mean residence time were obtained (equations were found appropriate).

Sponsor's Results:
The discussion is based on results of the assay by Squibb.

A mean of 74% of the administered dose of aztreonam was excreted unchanged in the urine during the 24 h after administration. An additional 2.6% was excreted as the open-ring metabolite.

The following table summarizes mean pharmacokinetic parameters ir CF patients 8-18 yrs of age:

	$t_{\frac{1}{2}}$ hr	MRT hr	V_{dss} ml/kg	Cl_s ml/min/kg	Cl_s ml/min/m²	Ue % of dose
Mean	1.54	1.80	246	2.46	71	74
SEM	0.14	0.19	17	0.22	5	3
N	10	10	10	10	10	9

Table 20 (reproduced in Appendix VI) contains individual subject pharmacokinetic parameters. Serum aztreonam clearance did not correlate significantly with creatinine clearance (Table 21, reproduced in Appendix VI). A similar phenomenon has been observed with gentamicin.

Table 22 lists pharmacokinetic parameters in patients with cystic fibrosis (CF) and for children and adults who did not have cystic fibrosis. Compared to non-CF children 2-12 yrs, and to adults, CF patients have an increased serum clearance (when expressed on a BSA basis) and a shortened serum half-life.

Clearance of aztreonam, expressed on a body weight basis, was similar for CF patients compared to non-CF children, but significantly greater than that reported for normal adults. These are not unexpected findings since CF patients have enhanced elimination of many other antibiotics.

Conclusion:
Study #18554-62 is acceptable in that it describes the pharmacokinetics of aztreonam in cystic fibrosis patients 10-18 years, after a single, 30 mg/kg IV dose infused over 3 minutes.

Comments:
12. Both, the sponsor and the clinical investigator assayed the

serum samples using different methods. The sponsor indicated that the assay results at the two institutions were generally in good agreement (correlation coefficients for serum and urine exceeded 0.98). Serum aztreonam concentrations measured by the clinical investigator (included in Appendix C of the report) indicate that the assay was less sensitive. Values also reflect higher variability than the sponsor's assay. Therefore, values obtained by the clinical investigator will not be considered in this review.

13. Based on the estimated clearance (per kg basis) observed in CF patients (8-18 yrs) and on the recommended therapy for adults with life-threatening infections (2 g aztreonam q 6 h, approximately 30 mg/kg for a normal 70 kg adult), the sponsor suggests a dose of 50 mg/kg given every 6 h to treat CF patients 8-18 yrs with life-threatening infections should be studied further. (The equations used by the sponsor are similar to those included in study #18554-32 (on page 5 of this review), substituting CF patient clearance in place of the pediatric group age clearance).

14. Based on BSA, the appropriate dose for CF patients (8-18 yrs) would be 1896 mg/sq.m (not 1635 mg/sq.m) given every 6 hours. The sponsor indicated that dosing on a BSA basis might be appropriate for CF patients who fall in the age range investigated in this study (8-18 yrs) but who are significantly outside the normal height and/or weight range for their age.

I. Responses to additional information requested on May 3, 1988

Comment #1: Provide estimates of steady state-serum aztreonam levels for each age and dose group evaluated in the major pediatric studies of aztreonam. Separate estimates should be made for patients with the highest AUC, the lowest AUC, and for the mean pharmacokinetic parameters. Projected steady-state levels should be plotted as a graph showing the MIC_{90} for organisms of interest.

Response to comment #1: The estimates of steady-state serum aztreonam levels for each age and dose group evaluated in the major pediatric studies are provided in the attached table (Section 1 of the attachments) entitled "Mean Aztreonam Serum Concentration (ug/ml)". Four graphs of projected steady-state levels showing the MIC for organisms of interests are also attached.

Comment:
15. The sponsor's response to Mr. Hunt's comment #1 is acceptable. Table and figures have been reproduced in Appendix VII of this review. Mean projected steady-state aztreonam serum concentrations remain above the MIC_{90} of relevant pathogens for the 6 hour dosing interval.

Comment #2: The phase III study for each age group should be identified as the biopharmacokinetics division did not receive any Phase [redacted] data.

Response to comment #2: This information is contained in Section 2 of the attachments in the report entitled "Steady-state Serum Aztreonam Levels in Pediatric Patients." The report includes a summary of results of our major pediatric studies: Protocols 18,554-32, -32 Addendum A, -52, -52 Addendum A, -58 Addendum A, -58 Addendum D, and -62. Complete results of these studies have been appended for your convenience.

Comment:
16. The sponsor's response to Mr. Hunt's comment #2 is acceptable. It should be noted that, according to the information provided, all studies reviewed in the present document (vis, #18,554-32, -32A, -52, -52A, -58A, -58D and -62) were Phase III studies. Also, note that these studies (except for studies -58A and -58D) were all single dose studies. The sponsor has included, for each single dose study, projected steady-state levels based on the results obtained after single dose administration of aztreonam.

Comment #3: For 2-12 year old patients, the projected steady-state serum level for a 50 mg/kg dose should be made based on the single dose studies of 30 and 50 mg/kg. The two projections should be compared.

Response to comment #3: This information is provided in the attached table and graphs (Section 3) entitled, "18,554-32: Mean Aztreonam Serum Concentration (ug/ml)."
(Table and graphs are included in Appendix IX of this review.)

Comment:
17. The sponsor's response to Mr. Hunt's comment #3 is acceptable. Note that projected mean steady-state levels of aztreonam in children 2-12 years remain above the MIC_{90} of relevant pathogens during the 6 hour dosing interval. Results based on the 30 mg/kg dose were similar to those based on the 50 mg/kg dose.

Comment #4: For each meningitis study, CSF levels should be plotted on a graph that shows the MIC's for relevant pathogens.

Response to comment #4: Graphs showing the MICs for relevant pathogens for each meningitis study are attached in Section 4 of the attachments.

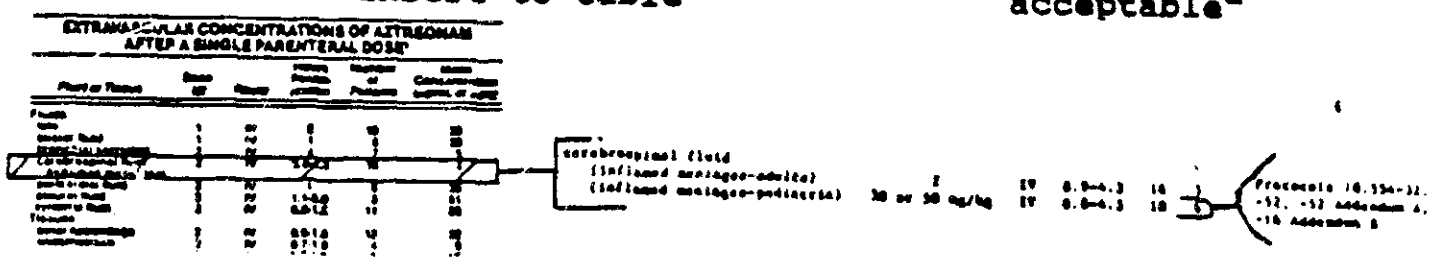
Comment:
18. The sponsor's response to Mr. Hunt's comment #4 is acceptable. In patients for which CSF aztreonam concentrations were obtained, CSF concentrations were above the MIC_{90} of E.

coli, N. meningitidis and H. influenza during the time periods studied (refer to graphs reproduced in Appendix X). In some cases, CSF levels were below the MIC_{90} of P. aeruginosa.

J. Proposed labeling (Package Insert) revisions:

A copy of the proposed package insert is found in Appendix XI attached to this review. Recommendations itemized by page follow:

Insert page(s)	Revision proposed	Recommendation
1, 5, 10, 11	none	-
2, 8	add subsection heading	acceptable
3	insert to table	acceptable[a]


EXTRACELLULAR CONCENTRATIONS OF AZTREONAM AFTER A SINGLE PARENTERAL DOSE[a]

[a]Data pertaining to CSF levels are accurate based on studies #18554-32, -52 and -52A.

6, 7	insert to pediatric use and insert to Indications and Usage	see comment #19
4	pharmacokinetics in pediatric patients	acceptable with modifications (see comment #20)
9	recommended doses in pediatric patients	see comment #21

Comments:
19. The revisions proposed in pages 6 and 7 of the Package Insert are clinical in nature and pertain to the Medical Officer.

20. Data presented in the Package Insert table on page 4 are accurate based on results of the indicated studies. Data for the 50 mg/kg dose (third line in the body of the table) should be aligned with the 2-12 yr old group so that it clearly indicates that the data arises from that specific group of patients. Also, it is recommended that for the second group (<1 week, >2.5 kg or

1 week-2 years) an additional line be inserted to include the results of studies #18554-52 and -52-A obtained after a dose of 50 mg/kg infused over 30 minutes.

Sponsor's proposed revisions to page 4 of insert:

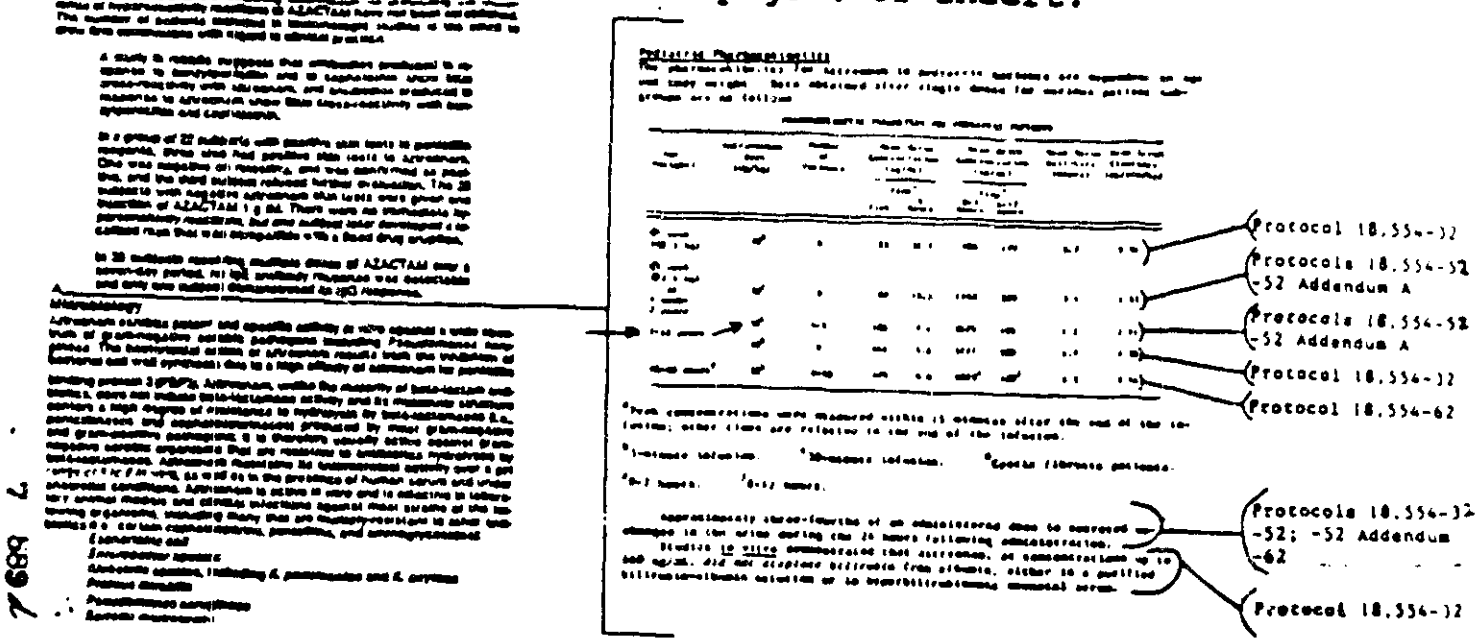


Table of pharmacokinetic parameters after recommended revisions are incorporated:

Pediatric Pharmacokinetics
The pharmacokinetics of aztreonam in pediatric patients are dependent on age and body weight. Data obtained after single doses for various patient subgroups are as follows:

PHARMACOKINETIC PARAMETERS FOR PEDIATRIC PATIENTS

Age (weight)	Intravenous Dose (mg/kg)	Number of Patients	Mean Serum Concentration (ug/ml) Time[a] Peak	Mean Serum Concentration (ug/ml) Time[a] 6 Hours	Mean Urine Concentration (ug/ml) Time[a] 0-3 Hours	Mean Urine Concentration (ug/ml) Time[a] 6-12 Hours	Mean Serum Half-Life (hours)	Mean Serum Clearance (mL/min/kg)
<1 week (<2.5 kg)	30[b]	6	83	31.7	786	179	5.7	0.94
<1 week (>2.5 kg) or 1 week-2 years	50[c]	5-6	211	26.9	2824	502	3.0	1.71
	30[c]	6	82	15.3	1142	329	3.4	1.53
2-12 years	50[c]	4-5	186	7.4	3675	196	1.2	2.51
	30[b]	6	141	5.8	3727	506	1.7	2.50
10-18 years[d]	30[b]	8-10	175	5.6	5827[e]	122[f]	1.5	2.46

[a]Peak concentrations were measured within 15 minutes after the end of the infusion; other times are relative to the end of the infusion.

[b]3-minute infusion. [c]30-minute infusion. [d]Cystic fibrosis patients.

[e]0-2 hours. [f]8-12 hours.

Based on the results of studies #18554-32, -52, -52A and -62, the first sentence after the table (page 4 of 11) should be changed to read:

> Approximately 60% of an administered dose is excreted unchanged in the urine during the 24 hours following administration. In cystic fibrosis patients (10-18 years) this value is increased to approximately 75%.

The second paragraph after the table should be evaluated by a biochemist (see comment #5).

21. The following revisions are proposed by the sponsor:

p. 9 of 11 pp.
AZACTAM
April 30, 1987

CURRENT INSERT

PROPOSED REVISIONS

weeks; some infections such as osteomyelitis may require therapy for four to six weeks.) Protocol 18,554-58

Pediatric
The usual dosage for patients older than one month is 30 mg/kg every six to eight hours. For severe infections in patients two years or older, 50 mg/kg every six to eight hours is recommended.
The recommended dose for all patients in the treatment of infections due to Ps. aeruginosa is 50 mg/kg every six to eight hours.
The maximum daily pediatric dose should not exceed the maximum recommended dose for adults.

Protocols 18,554-32; -52; -52 Addendum A; -62; -16A Addendum B

It is recommended that the section: **DOSAGE AND ADMINISTRATION** Pediatric, should be revised as follows (or similar text):

> Pediatric
> The usual dosage for patients older than one month with normal renal function is 30 mg/kg every six to eight hours administered as an intravenous infusion over 3 to 30 min. For severe infections in patients two years or older, 50 mg/kg every six to eight hours is recommended.
>
> The recommended dose for the treatment of infections due to Ps. aeruginosa in pediatric patients older than one month with normal renal function is 50 mg/kg every six to eight hours.

[redacted] maximum daily pediatric dose should not exceed [redacted] maximum recommended dose for adults (8 g per [redacted]

22. Under **RENAL IMPAIRMENT** (page 9 of package insert) separate subsections for Adults and Pediatric should be included. Under Pediatric the following is suggested:

> Pediatric
> The safety, effectiveness and pharmacokinetics of aztreonam in pediatric patients with impaired renal function have not yet been established.

General comments:
1. The results of study 18554-58-A give some evidance to support the linearity of aztreonam kinetics. Although only six patients were studied (ages 6-8 months and 2-12 years), there is no apparent accumulation of drug upon multiple dosing. Actual aztreonam serum levels at each time point agreed well with predicted levels (Appendix VIII, Table 1) based on single dose data. In children 2-12 years, predicted aztreonam levels for a single dose of 50 mg/kg, based on a single dose of 30 mg/kg agreed well with actual data after a single dose of 50 mg/kg (Appendix VII, page 1). In addition, pharmacokinetic parameters estimated after multiple dosing agreed well with values obtained after single dosing.

2. The data reviewed in this document supports the assumption of linear kinetics of aztreonam in pediatric patients (with normal renal function). However, the data is limited and not all pediatric groups were included in the multiple dose study.

3. The studies reviewed herein support the intravenous use of aztreonam in the populations studied.

Recommendations:
As requested by the Division of Anti-Infective Drug Products (HFD-520), the Division of Biopharmaceutics has reviewed Amendments S-006 (Feb. 23, 1987) and S-008 (May 18,1987), the Response to FDA Request for Information (October 12, 1988), and the revisions to Package Insert, which were submitted in support of NDA 50-580.

1. The current submission contains multiple dose data on only 6 pediatric patients, 4 patients with ages 11-12 years and 2 patients with ages 0.5 to 0.67 years, who received a dose of 30 mg/kg administered as 30-min infusions every 6 or 8 hours.
The doses recommended in the label were determined based on clearance estimates for each pediatric group obtained from single

dose studies. Because of the limited data available on multiple dosing, it would be advisable to evaluate, in a small number of pediatric patients of each age group, the recommended doses in multiple dose regimens in order to compare the actual steady state levels with the projected levels based on single doses. The data gathered will permit confirmation of the linearity in aztreonam kinetics in pediatric patients and will insure the safety of aztreonam at the recommended doses. This Division would welcome the opportunity to review the protocol for such study prior to its initiation.

2. Once the revisions to the proposed Package Insert are incorporated, the sponsor is required to re-submit such document for final review. It should be noted that the sponsor has not included a proposed dose regimen for use in cystic fibrosis patients.

These recommendations, deficiency #1 (page 13), comments #4 (p.7), #8 (p.11), #11 (p.14), #14 (p.16), #20 (p.18-20), #21 (p.20-21), #22 (p.21), and General comments #2 and #3 (p.21) should be forwarded to the sponsor.

Iraida González
Iraida Gonzalez, Ph.D.
Division of Biopharmaceutics

RD Initialed by John P. Hunt 11/28/89
FT Initialed by CT Viswanathan, Ph.D. JPH 11/29/89 CTV

cc: NDA 50-580 Orig., HFD-520 (2), HFD-426 (Viswanathan, Hunt, Gonzalez), HFD-344(Turner), Drug, Chron. and FOI files.

IG:CPQ:WP:102789

APPENDIX I

SQUIBB RESEARCH AND DEVELOPMENT
DIVISION OF MEDICAL AFFAIRS

TABLE 1[a]

18554-29

Number of Patients	0.5-Hr Serum Conc. µg/ml	Time of Tap, hr	Serum Conc. at Time of Tap, µg/ml	CSF Conc. µg/ml
Normal Meninges				
6	145±16	1.18±0.19	97.7±18.2	0.50±0.20
5	140±43	4.09±0.25	35.3±12.9	0.94±0.23
3	150±20	4.75±0.11	26.9±4.0	1.03±0.20
5	137±22	5.92±0.17	14.9±8.1	0.67±0.26
5	125±15	8.03±0.22	8.46±1.32	0.94±0.60
1	130	9.00	3.04	1.19
Inflamed Meninges				
5	126±18	1.09±0.18	88.4±21.5	1.98±3.44
1	139	2.17	54.7	1.98
3	112±27	4.15±0.16	18.0±7.2	3.22±2.99

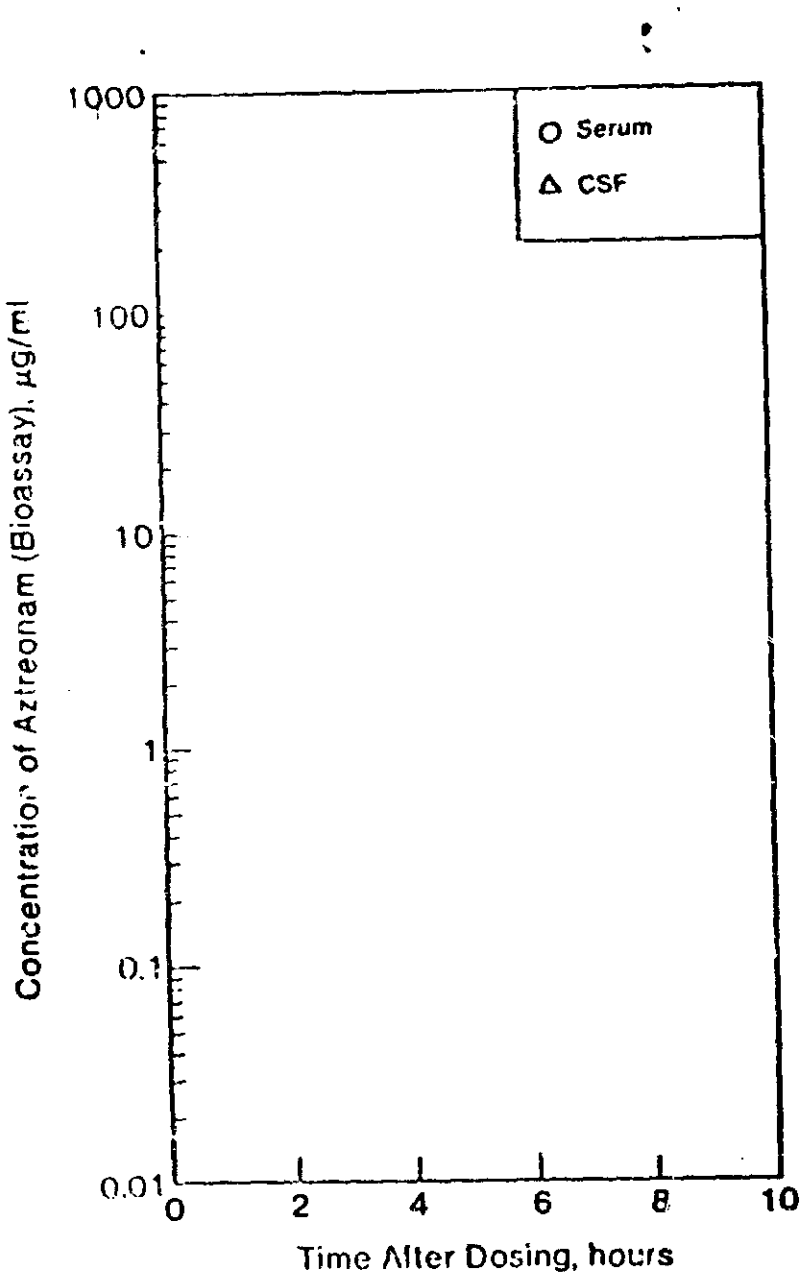
[a]Values are mean ± SD, (range); concentrations were determined by microbiological assay.

S-011

SQ 1924 9/74

FIGURE 5

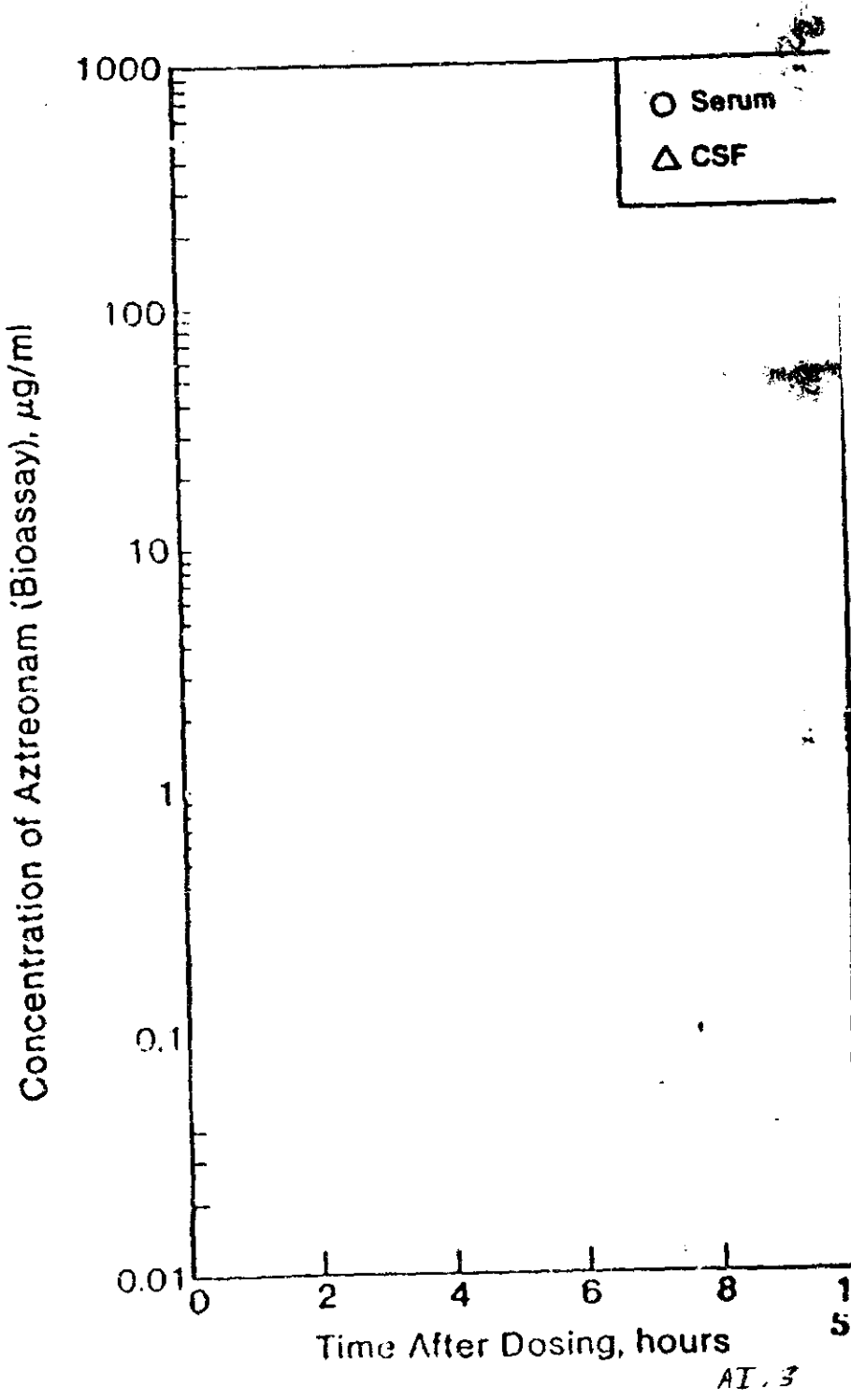
PENETRATION OF AZTREONAM INTO CEREBROSPINAL FLUID OF PATIENTS WITH NORMAL MENINGES AFTER A 2-GRAM INTRAVENOUS DOSE



AT.2

FIGURE 6

PENETRATION OF AZTREONAM INTO CEREBROSPINAL FLUID OF PATIENTS WITH INFLAMED MENINGES AFTER A 2-GRAM INTRAVENOUS DOSE



AT.3

Protocol No. 18554-51
(Revised 8/22/83)

TABLE 1

HPLC AZTREONAM ASSAY RESULTS (mcg/ml)

Patient Number		SERUM TIME (hours) 0	0.5	1	2	4	8	CSF TIME (hours) 0	0.5	1	2	4	8
006	I												
007	I												
008	I												
013	I												
014	I												
015	I												
017	I												
MEAN (I)		0	108		44.1	17.8	-	-	-	-	3.44	6.24	-
009	E												
010	E												
011	E												
[b]012	E												
MEAN (E)		-	78.8	-	33.2	-	-	-	-	1.36	1.92	2.14	3.31
OVERALL MEAN[6]			101	-	41.4	14.8				1.36	2.79	4.60	3.31

I - Included; E - Excluded

a - Bioassay result - no HPLC assay done

b - Patient 012 had a positive pre-dose serum aztreonam concentration, probably due to mislabeling of sample tubes. Serum data for this patient is excluded from the MEAN.

FIGURE 1 Protocol No. 18554-51

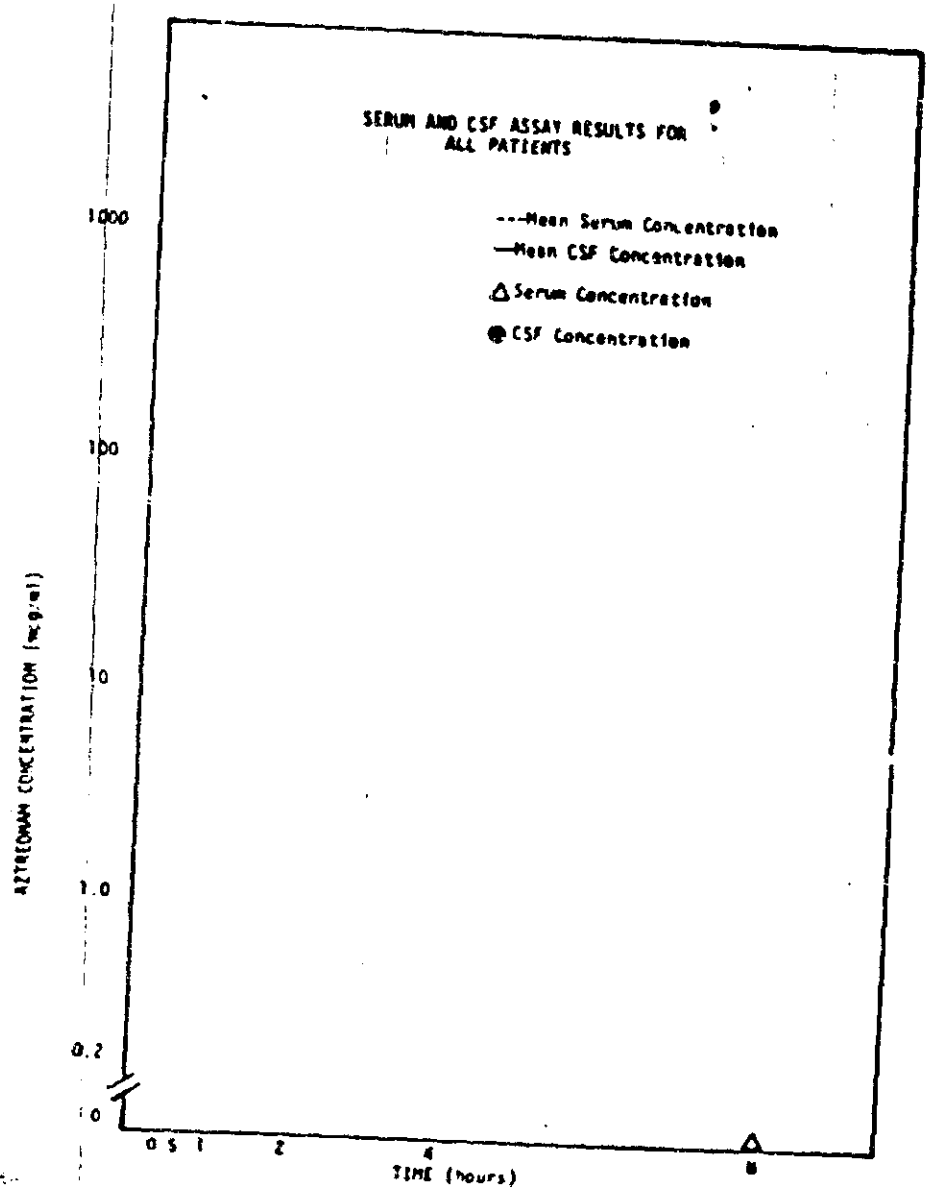
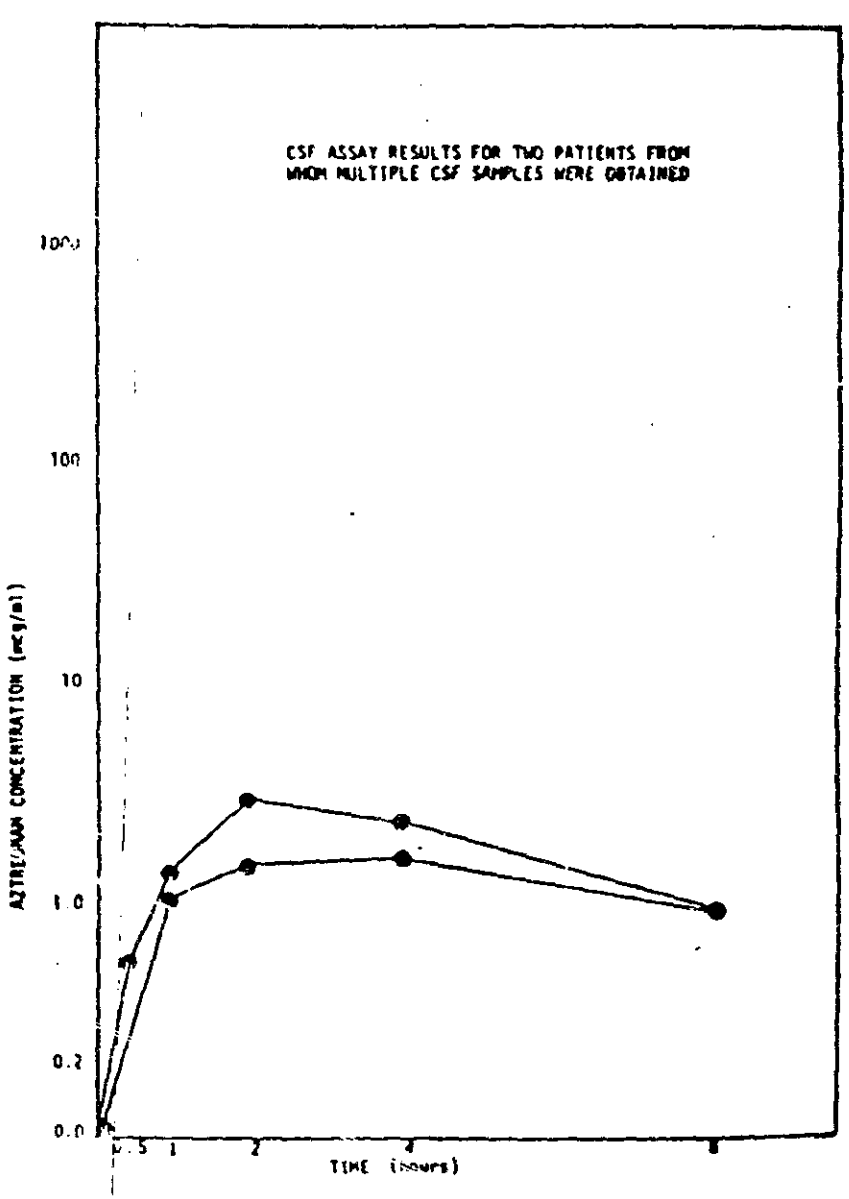


FIGURE 2 Protocol No. 18554-51



Appendix II

APPENDIX II

18554-32
NDA 324

Mean ± SD concentrations (μg/ml) of aztreonam in serum are given in the following table. SQ 26,992 (the open-beta-lactam-ring hydrolysis product of aztreonam) could not be reliably quantitated in serum of study patients. Mean levels of aztreonam at 15 minutes after infusion were approximately 100 μg/ml, and at 6 hours were 6 to 32 μg/ml, depending upon the age and weight group.

Age	Time After Dosing, hr				Number of Patients
	0.25	1	3	6	
Newborns					
<1 wk, <2.5 kg	83.0 ±52.4	71.9 ±10.1	46.2 ±7.0	31.7 ±0.1	6
<1 wk, >2.5 kg	97.8 ±12.3	75.4 ±12.7	45.4 ±10.9	17.6 ±5.5	6
1 wk - 1 mo	83.7 ±31.2	64.0 ±8.8	38.0 ±7.5	14.1 ±0.1	5
Infants					
>1 mo - 2 yr	115.5 ±16.1	103.6 ±86.1	24.5 ±10.4	11.6 ±13.2	6
Children					
>2 yr - 12 yr	140.8 ±112.2	54.4 ±13.9	18.1 ±5.0	5.8 ±3.0	6

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Abstract Page 5

SUMMARY OF PHARMACOKINETIC PARAMETERS[a]

Age Category	N	$t_{\frac{1}{2}}$ hr	MRT hr	$V_{D,SS}$ liters/kg	CL_S ml/min/kg
1. Newborns					
a. <1 wk, <2500 gm	6	5.71 ± 1.63 4.75	8.05 ± 2.32 6.69	0.36 ± 0.04	0.94 ± 0.14
b. <1 wk, >2500 gm	6	2.56 ± 0.20 2.52[A2]	3.32 ± 0.36 3.19[A2]	0.26 ± 0.02	1.41 ± 0.15
c. 1 wk - 1 mo	5	2.43 ± 0.35 2.34[A2]	3.15 ± 0.46 3.04[A2]	0.30 ± 0.02	1.68[A1] ± 0.16
2. Infants >1 mo - 2 yr	6[b]	1.70 ± 0.16 1.66[A3]	1.97 ± 0.20 1.92[A3]	0.20[A1] ± 0.03	1.87[A2] ± 0.31
3. Children >2 yr - 12 yr	5[c]	1.67 ± 0.21 1.62[A3]	1.93 ± 0.33 1.84[A3,B1]	0.29 ± 0.07	2.50 ± 0.15 A3, B2, C1, D1

[a]Values are arithmetic mean ± SEM, followed by geometric mean for $t_{\frac{1}{2}}$ and MRT. Results of statistical analysis are given as follows:
A: different from newborns <1 wk, <2500 gm.
B: different from newborns <1 wk, >2500 gm.
C: different from newborns 1 wk - 1 mo.
D: different from infants.
1) p <0.05
2) p <0.01
3) p <0.002

[b]Patient 6, who received multiple doses of aztreonam prior to this pharmacokinetic study, was omitted from this statistical analysis.

[illegible] half-life of 14.4 hr and MRT of 9.90 hr in the presence of normal renal function, and

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TABLE 24

PATIENTS GIVING CSF SPECIMENS FOR ASSAY OF AZTREONAM BY AGE CATEGORY

Pt. No.	Diagnosis	CSF Clinical Tests: RBC no/mm³	WBC & Diff[a] no/mm³	Glucose mg/dl	Protein mg/dl	Time of Tap Post Dose, hr	Conc. Az[b] Ser	Conc. Az[b] CSF
1a. Newborns, <1 wk, <2500 gm:			none					
1b. Newborns, <1 wk, >2500 gm:			none					
1c. Newborns, 1 wk - 1 mo:								
14	*Streptococcous pneumoniae* meningitis	12	190(N30,M70)	29	130	0.75	60.6 (1 hr)	2.33
24	*E. coli* meningitis	200	158(N63,M37)	12	240	1.33	68.4 (1 hr)	13.3
2. Infants, >1 mo - 2 yr:								
6[c]	*Enterobacter* ventriculitis	15	22(N 22)	17	35	2 3	49.2	9.32 10.9
9	*Hemophilus influenzae* meningitis	49	821(N84,M16)	49	61	4.33	22.9 (3 hr)	2.07
11	*Streptococcus pneumoniae* meningitis	52	412(N81,M19)	67	qns[e]	0.75	69.8 (1 hr)	20.8

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TABLE 24 (cont'd)

PATIENTS GIVING CSF SPECIMENS FOR ASSAY OF AZTREONAM BY AGE CATEGORY

Pt. No.	Diagnosis	CSF Clinical Tests: RBC no/mm³	WBC & Diff[a] no/mm³	Glucose mg/dl	Protein mg/dl	Time of Tap Post Dose, hr	Conc. Az[b] Ser	Conc. Az[b] CSF
3. Children, >2 yr - 12 yr:								
15	*Streptococcus pneumoniae* meningitis	3185[d]	23790(N94,M6)	22	248	1.08	44.5	3.4

[a] ug/ml, by high-pressure liquid chromatography assay.

[b] N = neutrophils, M = mononuclear cells.

[c] Studied after multiple doses; not evaluable in terms of the procedure specified in the protocol.

[d] Exceeded 500/mm³, suggesting a traumatic tap.

[e] qns = quantity not sufficient.

TABLE 25

Protocol 18554-3

PHARMACOKINETIC PARAMETERS FOR PEDIATRIC PATIENTS RECEIVING A 30 MG/KG, 3-MIN INTRAVENOUS INFUSION OF AZTREONAM BY AGE CATEGORY

Patient No.	$t_{\frac{1}{2}}$, hr	MRT, hr	$V_{D, SS}$ liters	$V_{D, SS}$ liters/kg	Urinary Excr. % of dose	CL_s ml/min	CL_s ml/min/kg	CL_r ml/min	CL_r ml/min/kg	CL_{nr} ml/min	CL_{nr} ml/min/kg
1a. Newborns, <1 wk, <2500 gm:											
19											
20											
21											
23											
27											
28											
1b. Newborns, <1 wk, >2500 gm:											
22											
25											
26											
29											
30											
31											

TABLE 25 (Cont'd)

Protocol 18554-

PHARMACOKINETIC PARAMETERS FOR PEDIATRIC PATIENTS RECEIVING A 30 MG/KG, 3-MIN INTRAVENOUS INFUSION OF AZTREONAM BY AGE CATEGORY

Patient No.	$t_{\frac{1}{2}}$, hr	MRT, hr	$V_{D, SS}$ liters	$V_{D, SS}$ liters/kg	Urinary Excr. % of dose	CL_s ml/min	CL_s ml/min/kg	CL_r ml/min	CL_r ml/min/kg	CL_{nr} ml/min	CL_{nr} ml/min/kg
1c. Newborns, >1 wk - 1 mo:											
7											
12											
14											
16											
24											
2. Infants, >1 mo - 2 yr:											
1											
3											
5											
6											
9											
10											
11											

Protocol 18554-32

TABLE 25 (Cont'd)

PHARMACOKINETIC PARAMETERS FOR PEDIATRIC PATIENTS RECEIVING A 30 MG/KG, 3-MIN INTRAVENOUS INFUSION OF AZTREONAM BY AGE CATEGORY

Patient No.	$t_{\frac{1}{2}}$, hr	MRT, hr	$V_{D, SS}$ liters	$V_{D, SS}$ liters/kg	Urinary Excr. % of dose	CL_s ml/min	CL_s ml/min/kg	CL_r ml/min	CL_r ml/min/kg	CL_{nr} ml/min	CL_{nr} ml/min/kg
3. Children, >2 yr - 12 yr:											
4											
8											
13											
15											
17											
18											

[a]It is likely that there was an error in specimen handling or recording of urine volumes, to explain over 100% recovery of dose. CL_r and CL_{nr} are shown to illustrate the standard methods of data analysis for such a patient, but are also inaccurate.

Protocol 18554-32

TABLE 26
SUMMARY OF PHARMACOKINETIC PARAMETERS[a]

Age Category	N	$t_{\frac{1}{2}}$, hr	MRT, hr	$V_{D,SS}$, liters/kg	CL_s, ml/min/kg
1. Newborns					
a. < 1 wk, <2500 gm	6	5.71 ± 1.63 4.75	8.05 ± 2.32 6.69	0.36 ± 0.04	0.94 ± 0.14
b. < 1 wk, >2500 gm	6	2.56 ± 0.20 2.52^{A2}	3.32 ± 0.36 3.19^{A2}	0.26 ± 0.02	1.41 ± 0.15
c. 1 wk - 1 mo	5	2.43 ± 0.35 2.34^{A2}	3.15 ± 0.46 3.04^{A2}	0.30 ± 0.02	1.68^{A1} ± 0.16
2. Infants >1 mo - 2 yr	6[b]	1.70 ± 0.16 1.66^{A3}	1.97 ± 0.20 1.92^{A3}	0.20^{A1} ± 0.03	1.87^{A2} ± 0.31
3. Children >2 yr - 12 yr	5[c]	1.67 ± 0.21 1.62^{A3}	1.93 ± 0.33 $1.84^{A3,B1}$	0.29 ± 0.07	2.50 ± 0.15 A3, B2, C1, D1

Footnotes for Table 26

[a] Values are arithmetic mean ± SEM followed in some cases by geometric mean. Results of statistical analysis are given as follows:

A: different from newborns <1 wk, < 2500 gm.
B: different from newborns <1 wk, > 2500 gm.
C: different from newborns 1 wk - 1 mo.
D: different from infants.

1) $p < 0.05$
2) $p < 0.01$
3) $p < 0.001$

[b] Patient 6, who received multiple doses of aztreonam prior to this pharmacokinetic study, was omitted from this statistical analysis

[c] Patient 8 had an apparent half-life of 14.4 hr and MRT of 9.90 hr in the presence of normal renal function, and was omitted from this statistical analysis.

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TABLE 27

INTERPRETATION OF AGE DEPENDENCE OF SERUM CLEARANCE OF AZTREONAM

Age Category	Inulin Clearance[a] ml/min/m²	SA[f] m²	Wt kg	Inulin Clearance ml/min/kg
newborn <4d old	11	0.21[b]	3.0[b]	0.77
14d old	~20	0.25[c]	4.0[c]	1.25
infant 1 yr old	~70 (adult value)	0.39[d]	7.7[d]	3.54
children		0.68[e]	16.7[e]	2.85
adult	~70	1.73	70	1.73

[a] Values from Hilligoss, 1980.

[b] Mean of Patients 25, 26, 29, 30 & 31, newborns ≤4 days & >2.5 kg.

[c] Mean of Patients 7, 12, 14, 16, & 24, newborns 1 wk - 1 mo.

[d] Mean of Patients 1, 3, 5, 9, 10, & 11, infants 1 mo - 2 yr.

[e] Mean of Patients 4, 8, 13, 15, 17, & 18, children 2 yr - 12 yr.

[f] SA is body surface area.

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TABLE 28

INTERPRETATION OF AGE DEPENDENCE OF VOLUME OF DISTRIBUTION AT STEADY STATE

Age Category	Body Water[a]		
	Total Body Water (TBW)	Intracellular Water (ICW)	Extracellular Water (ECW)
premature newborns	85% BWt[b]		
full-term newborns	78% BWt	43% TBW or 34% BWT	57% TBW or 44% BWT
(1 yr ~ adult TBW)			
adults	60% BWt	68% TBW or 41% BWt	32% TBW or 19% BWt

[a] D.M. Hilligoss, 1980.

[b] BWT = Body weight

TABLE 30

CALCULATED AGE-RELATED ADJUSTMENTS IN DOSAGE REGIMEN
FOR AZTREONAM IN PEDIATRIC PATIENTS

Age Group	Cl_s ml/min/kg	Fixed Dose Interval Variable Dose			Variable Dose Interval Fixed Dose			
		$\frac{Cl_s}{1.5}$	$\frac{Cl_s}{1.5}\times 30$ mg/kg	$\frac{Cl_s}{1.5}\times 15$ mg/kg	$\frac{1.5}{Cl_s}$	$\frac{1.5}{Cl_s}\times 6$ hr	$\frac{1.5}{Cl_s}\times 8$ hr	$\frac{1.5}{Cl_s}\times$ hr
1a. Newborns <1 wk & <2.5 kg	0.94	0.627	18.8	9.4	1.60	9.6	12.8	19.1
1b. Newborns <1 wk & >2.5 kg	1.41	0.94	28.2	14.1	1.06	6.4	8.5	12.8
1c. Newborns 1 wk - 1 mo	1.68	1.12	33.6	16.8	0.89	5.4	7.1	10.7
2. Infants >1 mo - 2 yr	1.87	1.25	37.4	18.7	0.80	4.8	6.4	9.6
3. Children >2 yr - 12 yr	2.50	1.67	50	25	0.60	3.6	4.8	7.2
Adults	1.5	1	30	15	1	6	8	12

SQUIBB RESEARCH AND DEVELOPMENT
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Figure 1

Comparison of Serum Pharmacokinetics
of Aztreonam (30-mg/kg Dose, IV) in
Pediatric Patients and In Vitro Bacterial Susceptibilities

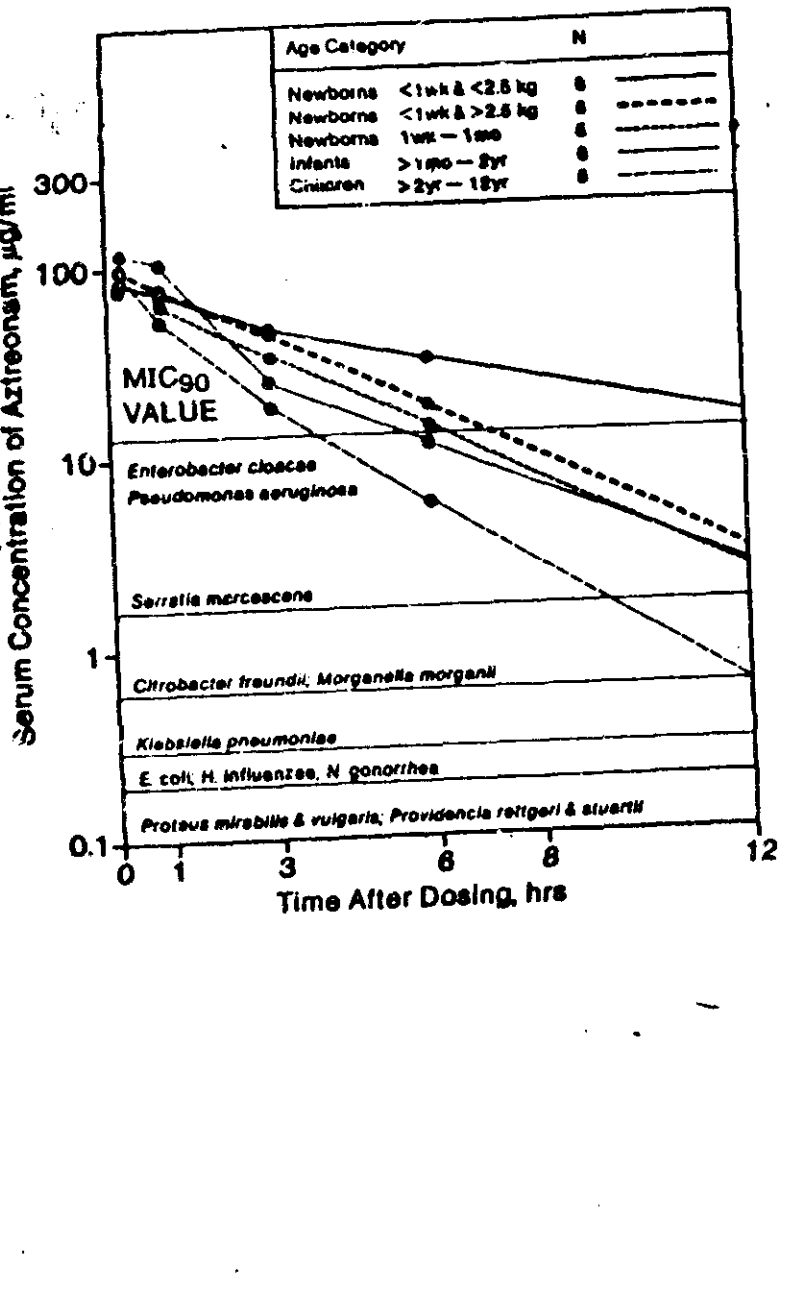

Figure 2

Comparison of Serum Pharmacokinetics
of Aztreonam (3-Min IV Infusion) in Pediatric
Patients (30-mg/kg Dose) and Healthy Adults (2-gm Dose)

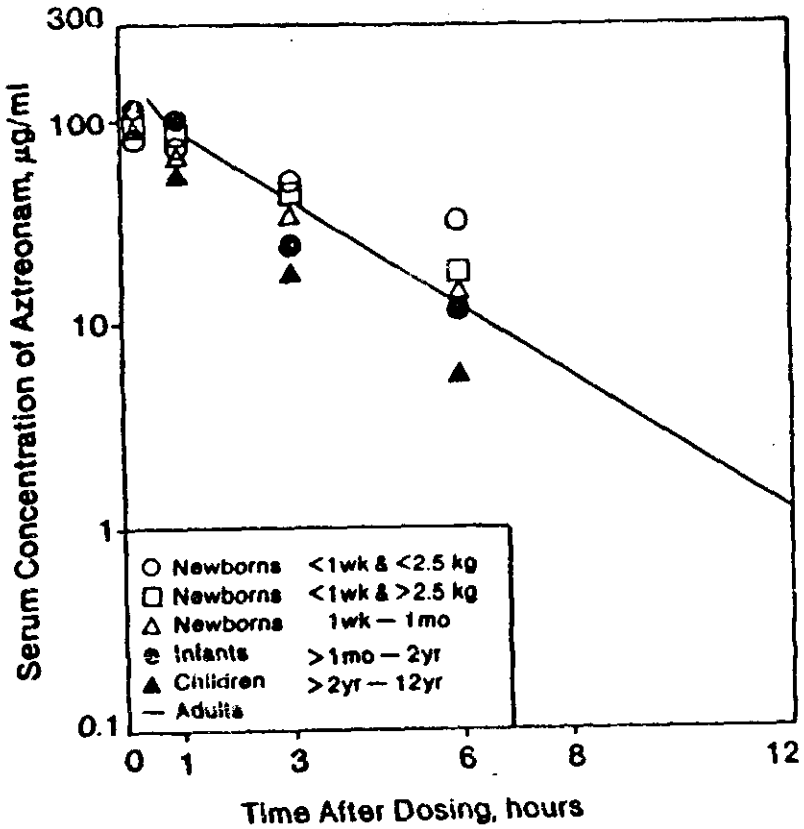

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Figure 3

Comparison of Estimated Serum Concentration of Aztreonam Based on Serum Inhibitory Power and Measured Serum Concentration of Aztreonam

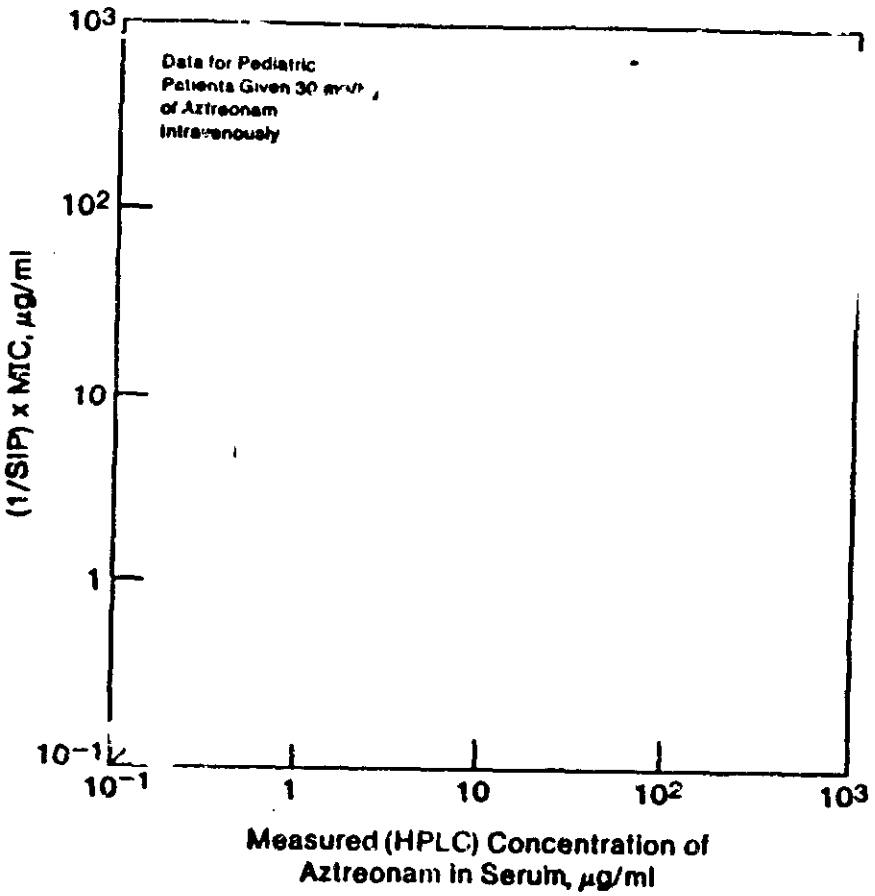
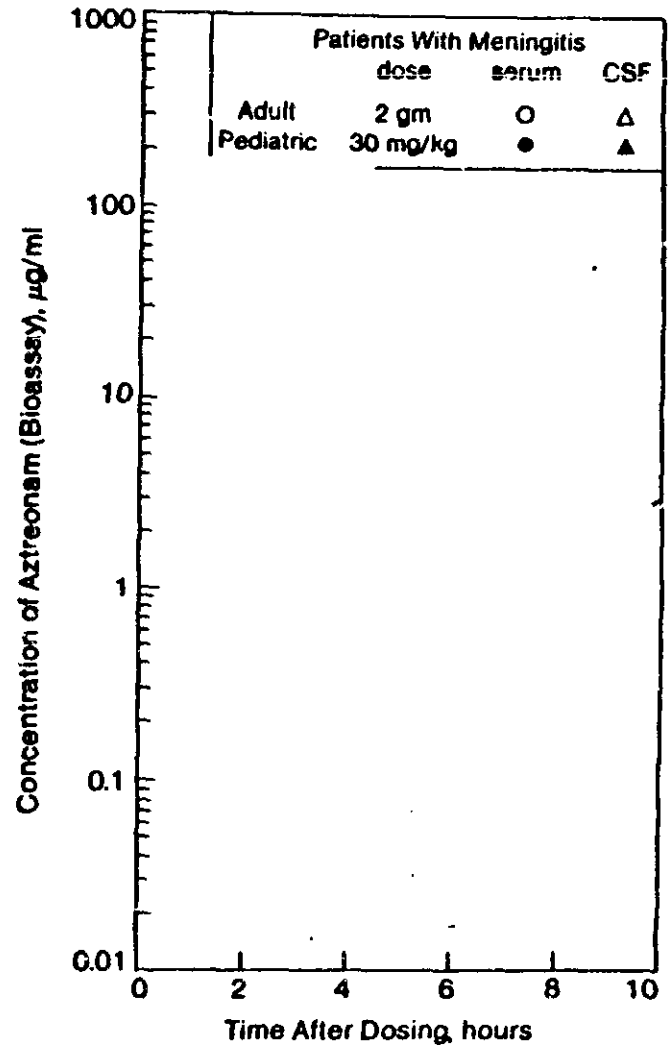


Figure 4



*Continuous curves, inserted as references for data points shown, were obtained from adult patients with normal meninges under Protocol 18554-29.

Protocol 18554-32
Addendum A

TABLE 16

SERUM AZTREONAM AND SQ 26,992 CONCENTRATIONS (μg/ml)

PATIENT	AZTREONAM					SQ 26,992				
	TIME, HRS									
	0.00	0.25	1.00	3.00	6.00	0.00	0.25	1.00	3.00	6.00
32										
33										
34										
35										
36										
37										
MEAN[c]		214.00	109.08	38.58	12.82		0.37	0.21	0.00	0.00
±S.E.M.		34.65	13.19	6.64	2.42		0.37	0.21	0.00	0.00

N.D. - Not determined

[a]Aztreonam in pre-dose serum was probably due to mistimed sample.

[b]Sample obtained at 0.5 hours.

[c]Sample obtained at 6.5 hours.

[d]These samples were probably mislabeled.

[e]Excluding Patient 35 whose samples were probably mislabeled.

Protocol 18554-32
Addendum A

TABLE 17

PHARMACOKINETIC PARAMETERS

PATIENT	$AUC_{0-\infty}$ (μg·hr/ml)	$AUMC_{0-\infty}$ (μg·hr²/ml)	$t_{\frac{1}{2}}$ (hr)	MRT (hr)	Serum Clearance (ml/min/kg)	Serum Clearance (ml/min/m²)	Renal Clearance (ml/min/kg)	V_{dss} (l/kg)	V_{dss} (l/m²)
32									
33									
34									
35[a]									
36									
37									
MEAN[b]	461.0	963.8	1.99	2.08	1.94	50.63	1.94	0.24	6.23
±SEM	63.2	170.9	0.23	0.21	0.24	1.00	0.53	0.04	0.60

[a]The 6-hour serum aztreonam concentration exceeded the 3-hour concentration for Patient 35, probably due to mislabeled samples. For the purpose of calculating these pharmacokinetic parameters, these assay results were interchanged.

[b]Excluding Patient 35 whose serum samples were probably mislabeled.

TABLE 18

URINE AZTREONAM AND SQ 26,992 CONCENTRATIONS (μg/ml)

PATIENT	AZTREONAM					SQ 26,992				
	TIME, HRS					TIME, HRS				
	PRE	0-3	3-6	6-12	12-24	PRE	0-3	3-6	6-12	12-24
32										
33										
34										
35										
36										
37										
Mean		3296.83	1660.40	358.00	34.79		0.00	34.95	44.00	36.60	35.95
±S.E.M.		994.19	669.00	94.92	12.90		0.00	9.92	12.70	5.55	7.62

TABLE 19

URINARY EXCRETION OF AZTREONAM IN PATIENTS
PROVIDING CUMULATIVE URINE COLLECTIONS

PATIENT	COLLECTION TIME (hr)	CONC. (μg/ml)	VOL. (ml)	AMOUNT (mg)	CUM. AMT. (mg)	CUM. AMT. % OF DOSE (%)
34						
Dose = 650 mg						
36						
Dose = 780 mg						
37						
Dose = 3000 mg						

TABLE 20

MEAN PHARMACOKINETIC PARAMETERS FOR PATIENTS
GIVEN 50 OR 30 MG/KG DOSES OF AZTREONAM[a]

Aztreonam Dose (mg/kg)	$t_{\frac{1}{2}}$ (hr)	MRT (hr)	Serum Clearance ml/min/kg	Serum Clearance ml/min/m²	V_{dss} l/kg
50[b]	1.99 ± 0.23	2.08 ± 0.21	1.94 ± 0.24	51 ± 1	0.24 ± 0.04
30[c]	1.67 ± 0.21	1.93 ± 0.33	2.50 ± 0.15	53 ± 4	0.29 ± 0.07

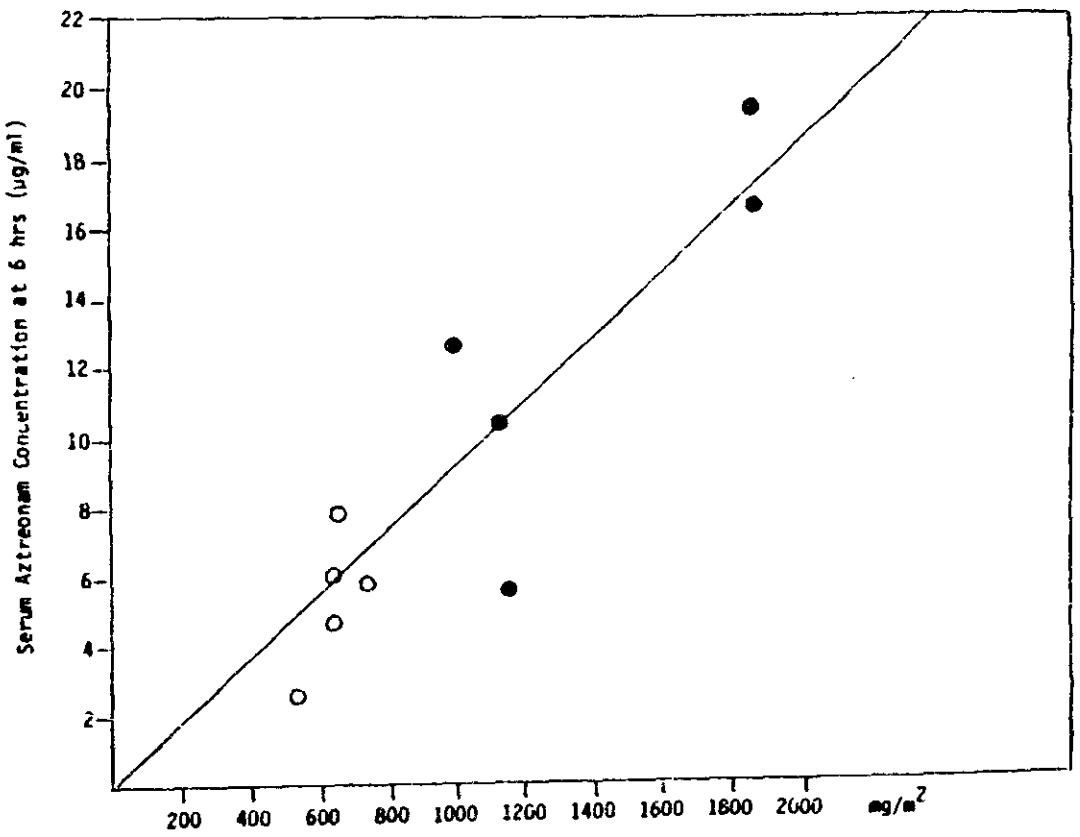
[a]Values given ± S.E.M.

[b]Patients of this study (Protocol 18554-32, Addendum A)

[c]Patients of Protocol 18554-32

Figure 2

Serum Aztreonam Concentration at 6 Hours as a Function of
Administered Dose



APPENDIX III

Protocol 18554-52 and 52A

TABLE 20

SERUM AZTREONAM CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Length of Infusion (hr)	Time (hrs) - Relative To The Beginning Of The Infusion			
				0.50	1.50	3.50	6.50
Age <1 wk and Wt <2500 gm	50	16[xx]					
		17*					
		19					
Age <1 wk and weight >2500 gm or Age 1 wk-2 yrs	30	2					
		4					
		5					
		6					
		7*					
		8					
		23					
		MEAN		82.1	66.4	31.4	15.3
		SEM		17.5	5.0	2.5	2.6
	50	9*					
		10					
		11					
		13					
		14					
		15					
		20[d]					
		MEAN		210.8	96.4	49.9	26.9
		SEM		38.0	20.8	14.6	8.8

*Not included in mean values.
NA-Sample could not be assayed because of technical difficulties.
[a]Time = 4.00 hr
[b]Time = 4.01 hr
[c]Time = 7.00 hr
[d]Pre-dose azt. conc. = 2.66 for this patient.
[xx]Pre-dose azt. conc. = 1.95 for this patient.

NOTE: Except for patients 16 and 20 all pre-dose values were 0.

TABLE 20
(continued)

SERUM AZTREONAM CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Length of Infusion (hr)	Time (hrs) - Relative To The Beginning Of Infusion			
				0.50	1.50	3.50	6.50
Age 2-12 yrs	30	1[d]	0.50	115.0	39.9	10.3	ND
	50	12					
		18*					
		21					
		22					
		24					
		26					
		MEAN		186.0	72.1	26.7	7.4
		SEM		24.8	12.3	3.9	0.9

[d]Additional sample taken at 1 hr contained ?? 0 µg/ml.
[e]Time = 4.58 hr
*Not included in mean values
ND-Not Determined

APPENDIX III

TABLE 21

URINE AZTREONAM CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Time (hrs) Relative To The Beginning of Infusion 0-3.5	3.5-6.5	6.5-12.5	12.5-24.5	(Pre-Dose Conc.)**
Age <1 Wk Wt <2500 gm	50	16					
		17*					
		19					
Age <1 Wk- and Weight >2500 gm or Age 1 Wk- 2 Yrs	30	2					
		4					
		5					
		6					
		7*					
		8					
		23					
		MEAN	1141.7	665.7	329.1	82.10	
		SEM	296.4	281.1	118.1	45.08	
	50	9*					
		10					
		11					
		13					
		14					
		15					
		20					
		MEAN	2823.6	1568.6	501.7	199.30	
		SEM	1694.6	722.3	78.9	116.00	

*Not included in mean values.
**All pre-dose samples except those listed were 0
***Probably a mislabeled post-dose sample
NS-No sample obtained

TABLE 21
(continued)

URINE AZTREONAM CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Time (hrs) Relative To The Beginning Of Infusion 0-3.5	3.5-6.5	6.5-12.5	12.5-24.5	(Pre-Dose Conc.)****
Age 2-12 yrs	30	1	NS	2490**	NS	66.40***	
	50	12					
		18*					
		21					
		22					
		24					
		26					
		MEAN	3675.0	3334.6	195.9	412.99	
		SEM	2271.1	1167.3	98.6	386.02	

ND-Not determined.
NS-No urine sample produced during this period.
*Not included in mean values.
**0-6.5 hr collection
***6.5-24.5 hr collection
****All pre-dose values, except those listed, were 0.

TABLE 22

CEREBROSPINAL FLUID (CSF) AZTREONAM AND SQ 26,992 CONCENTRATIONS

ATIENT GROUP	AZTREONAM DOSE (mg/kg)	PT NO	DIAGNOSES	CSF CLINICAL TESTS RBC /mm³	WBC & DIFF. /mm³	GLUCOSE mg/dl	PROTEIN mg/dl	TIME OF CSF SAMPLE (HR)*	SERUM CONCENTRATION (mcg/ml)	CSF AZTREONAM CONCENTRATION (mcg/ml) Azt.	SQ 26,992	COMMENT
ge <1 Wk and eight >2500 s or Age 1 k to 2 Yrs	30	8	H. Influenzae Meningitis	N.D.	N.D.	N.D.	N.D.	2.08	51 (1.58 hr)	2.46	1.28	
	30	7	H. Influenzae Meningitis	N.D.	N.D.	N.D	N.D	2.17	36 (1.75 hr)	21.00	0.00	Excluded Patient
	30	2	Pneumococcal Meningitis	36	56 (48 polys, 8 monos)	28	95	1.50	38.3 (3.5 hr)	4.23	0.00	
	50	20	H. Influenzae Meningitis	N.D.	N.D.	N.D.	N.D.	1.17	46.2 (1.5 hr)	2.87	0.00	
	50	11	H. Influenzae Meningitis	10	2120 (2134 polys, 186 monos)	50	74	1.67	70.6 (1.5 hr)	3.76	0.00	
	50	14	H. Influenzae Meningitis	2	2970 (2465 polys, 475 monos, 30 lymphs)	N.D.	N.D.	2.25	83.8 (1.83 hr)	8.70	0.00	
e 2-12 yrs	30	1	Meningococcal Meningitis and Sepsis	6	40 (39 polys, 1 mono)	112	29	3.00	39.9 (3.5 hr)	3.78	0.00	
	50	18	H. Influenzae Meningitis	367	508 (488 polys, 20 lymphs)	N.D.	N.D.	1.67	23.4 (1.5 hr)	1.70	0.00	Excluded Patient

N.D.-Not Determined
*Times are relative to the beginning of the aztreonam infusion

TABLE 23

SERUM SQ 26,992 CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Length of Infusion (hr)	Time (hrs) - Relative To The Beginning Of The Infusion			
				0.50	1.50	3.50	6.50
Age <1 Wk and Wt <2500 gm	50	16[xx]	0.55				
		17*	0.53				
		19	0.58				
Age <1 Wk and Wt >2500 gm or Age 1 Wk-2 Yrs	30	2	0.50				
		4	0.50				
		5	0.83				
		6	0.50				
		7*	0.75				
		8	0.50				
		23	0.50				
		MEAN		2.48	0.25	0.00	0.00
		SEM		2.14	0.25	0.00	0.00
	50	9*	0.50				
		10	0.52				
		11	0.50				
		13	0.50				
		14	0.50				
		15	0.50				
		20	0.50				
		MEAN		0.00	0.00	0.00	0.00
		SEM		0.00	0.00	0.00	0.00

*Not included in mean values.
NA- Sample could not be assayed because of technical difficulties.
[a]Time = 4.00 hr
[b]Time = 4.01 hr
[c]Time = 7.00 hr

NOTE: All pre-dose values were 0.

TABLE 23
(continued)

SERUM SQ 26,992 CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Length of Infusion (hr)	Time (hrs) - Relative To The Beginning Of The Infusion			
				0.50	1.50	3.50	6.50
Age 2-12 yrs	30	1[d]	0.50				
	50	12	0.50				
		18*	0.67				
		21	0.52				
		22	0.50				
		24	0.60				
		26	0.50				
		MEAN		1.02	0.62	0.19	0.20
		SEM		0.49	0.38	0.19	0.20

ND-Not determined
NA-Sample could not be assayed because of technical difficulties.
[d]Additional sample taken at 1 hr contained 3.08 µg/ml.
[e]Time = 4.58 hr
*Not included in mean values.

TABLE 24

URINE SQ 26 992 CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Time (hrs) Relative To The Beginning Of Infusion 0-3.5 hr	3.5 6.5 hr	6.5-12.5 hr	12.5-24.5 hr	(Pre-Dose Conc.)**
Age <1 wk Wt <2500 gm	50	16					
		17*					
		19					
Age <1 wk and Weight >2500 gm or Age 1 Wk-2 Yrs	?	2					
		4					
		5					
		6					
		7*					
		8					
		23					
		MEAN	11.75	12.64	17.93	15.75	
		SEM	1.67	2.30	6.17	3.83	
	50	9*					
		10					
		11					
		13					
		14					
		15					
		20					
		MEAN	24.30	23.48	28.45	17.74	
		SEM	17.84	11.07	13.55	7.88	

*Not included in mean values
**All pre-dose values, except those listed, were 0.
NS-No urine sample produced during this period.

TABLE 24
(continued)

URINE SQ 26,992 CONCENTRATIONS
(µg/ml)

Category	Dose (mg/kg)	Pat. No.	Time (hrs) Relative To The Beginning of Infusion 0-3.5 hr	3.5-6.5	6.5-12.5 hr	12.5 24.5 hr	(Pre-Dose Conc.)****
Age 2-12 yrs	30	1					
	50	12					
		18*					
		21					
		22					
		24					
		26					
		MEAN	32.20	67 03	50.53	32.77	
		SEM	20.86	24 39	14.73	11.59	

NA-Sample could not be assayed because of technical problems.
ND-Not determined
NS-No urine sample produced during this period.
*Not included in the mean values.
**0-6.5 hr collection
***6.5-24.5 hr collection
****All pre-dose values, except those listed, were 0.

TABLE 25

PHARMACOKINETIC PARAMETERS

CATEGORY	DOSE (mg/kg)	PAT. NO.	AUC (μg·hr/ml)	AUMC (μg·hr²/ml)	T½ (hr)	MRT (hr)	CL_p (ml/min)	CL_p (ml/min/kg)	CL_p (ml/min/m²)	CL_R (ml/min)	CL_R/kg (ml/min/kg)	V_{ss} (l)	V_{ss} (l/kg)
Age <1 Yr Wt <2500 gm	50	16											
		17											
		19											
Age <1 Yr and Wt >2500 gm or Age 1 Yr - 2 Yrs	30	2											
		4											
		5											
		6											
		7											
		8											
		23											
		MEAN	350.8	1656.7	3.370	6.500	9.76	1.53	27.5	5.69	0.812	1.967	0.367
		SEM	31.0	363.6	0.535	0.726	2.62	0.15	4.8	2.23	0.102	0.303	0.050

TABLE 25 (Continued)

PHARMACOKINETIC PARAMETERS

CATEGORY	DOSE (mg/kg)	PAT. NO.	AUC (μg·hr/ml)	AUMC (μg·hr²/ml)	T½ (hr)	MRT (hr)	CL_p (ml/min)	CL_p (ml/min/kg)	CL_p (ml/min/m²)	CL_R (ml/min)	CL_R/kg (ml/min/kg)	V_{ss} (l)	V_{ss} (l/kg)
Age <1 Yr and Wt >2500 gm or age 1 Yr - 2 Yrs	50	9											
		10											
		11											
		13											
		14											
		15											
		20											
		MEAN	615.9	2894.2	3.97	3.97	12.78	1.71	32.6	9.58	1.232	1.940	0.303
		SEM	119.8	1363.0	0.86	1.13	4.82	0.37	9.1	3.78	0.311	0.592	0.049
	Combined 30 or 50	MEAN			3.17	4.23	11.26	1.62	30.0	7.63	1.022	1.943	0.335
		SEM			0.49	0.91	2.65	0.19	4.8	2.17	0.168	0.317	0.033

TABLE 25 (Continued)

PHARMACOKINETIC PARAMETERS

CATEGORY	DOSE (mg/kg)	PAT. NO.	AUC (μg·hr/ml)	AUMC (μg·hr²/ml)	T½ (hr)	MRT (hr)	CL_p (ml/min)	CL_p (ml/min/kg)	CL_p (ml/min/m²)	CL_R (ml/min)	CL_R/kg (ml/min/kg)	V_{ss} (l)	V_{ss} (l/kg)
Age 2-12 Yrs	30	1	168.6	253.7	1.02	1.50	74.10	2.99	80.4	51.68	2.08	5.58	0.225
	50	12											
		18											
		21											
		22											
		24											
		26											
		MEAN	352.7	679.8	1.153	1.93	60.66	2.51	65.6	46.58	1.75	6.11	0.265
		SEM	46.5	89.6	0.172	0.09	11.08	0.39	2.16	6.22	0.24	1.17	0.043

Excluded - Excluded from calculation of Mean and SEM.

TABLE 26

COMPARISON OF AZTREONAM PHARMACOKINETIC PARAMETERS

Category	Protocol	Dose (mg)	$T_{\frac{1}{2}}$ (hr)	MRT (hr)	CL (ml/min/kg)	Vd$_{ss}$ (l/kg)
Age <1 Wk and Wt >2500 gm or Age 1 Wk-2 Yrs.	18554-32[a]	30	2.22	2.79	1.63	0.25
	This Study	30 or 50	3.17	4.23	1.62	0.34
Age 2-12 Yrs.	18554-32	30	1.67	1.93	2.50	0.29
	18554-32 Addendum A	50	1.99	2.08	1.94	0.24
	This Study	50	1.15	1.93	2.51	0.27

[a]Weighted averages calculated by combining data for patient subgroups reported in this study.

3 Pages

Purged

APPENDIX IV

Protocol 18554-58
Addendum A

TABLE 4

SERUM AZTREONAM CONCENTRATIONS (µg/ml)

PATIENT AGE	PAT.	TIME (RELATIVE TO THE END OF INFUSION) PRE		0.25		1.00		3.00		6.00	
		BEG.	END	BEG.	END	BEG.	END	BEG.	END	BEG.	END
	1										
	2										
11-12 yrs	4										
	5										
	MEAN	14.74	6.14	108.8	106.6	72.7	62.8	30.8	25.0	9.39	6.46
	SD	8.29	1.67	12.9	10.2	26.2	20.0	18.5	9.9	4.53	2.97
0.5-0.67 yrs	3										
	6										
	MEAN	1.89	5.48	52.1	32.2	42.1	58.0	30.4	39.4	2.91	12.75

Beg - Beginning of therapy
End - End of therapy

TABLE 5

PHARMACOKINETIC PARAMETERS FOR AZTREONAM

AGE	PAT.	C_{max} (µg/ml) BEG.	C_{max} (µg/ml) END	C_{min} (µg/ml) BEG.	C_{min} (µg/ml) END	AUC (hr·µg/ml) BEG.	AUC (hr·µg/ml) END	Cl_s (ml/min) BEG.	Cl_s (ml/min) END	Cl_s (ml/min/kg) BEG.	Cl_s (ml/min/kg) END	$t_{½}$ (hrs) BEG.	$t_{½}$ (hrs) END
	1												
	2												
11-12 yrs	4												
	5												
	MEAN	108.8	106.6	8.13	4.83	292.5	262.4	79.1	87.4	1.81	1.94	1.58	1.62
	SD	12.9	10.2	5.34	1.02	82.1	32.1	15.5	22.5	0.40	0.21	0.35	0.51
	N	4	4	4	4	4	4	4	4	4	4	4	4
0.5-0.67 yrs	3												
	6												
	MEAN	61.7	59.3	1.89	5.48	173.5	244.0	23.8	17.9	2.79	2.10	1.25	1.94
	N	2	2	2	2	2	2	2	2	2	2	2	2

BEG. - Beginning of therapy
END - End of therapy

TABLE 6

SERUM SQ 26,992 CONCENTRATIONS (µg/ml)

PATIENT AGE	PAT.	TIME (RELATIVE TO THE END OF INFUSION) PRE BEG.	PRE END	0.25 BEG.	0.25 END	1.00 BEG.	1.00 END	3.00 BEG.	3.00 END	6.00 BEG.	6.00 END
11-12 yrs	1										
	2										
	4										
	5										
	MEAN	1.53	0.79	1.91	1.55	1.46	1.21	2.58	0.79	1.46	0.69
	SD	1.08	0.93	1.37	1.07	1.02	0.82	2.68	0.93	0.98	0.81
0.5-0.67 yrs	3										
	6										
	MEAN	0.00	0.54	0.00	0.60	0.00	1.18	0.00	0.51	1.06	1.95

Beg - Beginning of therapy
End - End of therapy

APPENDIX V

PROTOCOL 18554-58 ADDENDUM D
ABSTRACT (CONTINUED)
PAGE 2

MEAN SERUM AZTREONAM CONCENTRATIONS
(µg/ml)

	PRE-INFUSION*		Time (Relative to End of Infusion) 0.0 HOURS**		0.5 HOURS		1 HOUR***		2 HOURS		4 HOURS***	8 HOURS
	DAY 1	DAY 3/4	DAY 1	DAY 3/4	DAY 1	DAY 3/4	DAY 1	DAY 1	DAY 3/4	DAY 1	DAY 1	DAY 3/4
MEAN	0	24.3	75.5	78.2	66.8	75.0	61.0	54.5	57.3	45.6	32.0	31.5
SEM	0	3.5	5.4	4.4	4.4	4.6	3.5	3.1	3.7	3.3	2.4	3.3
N	26	21	26	21	26	19	26	26	21	25	26	21

*10 minutes before infusion.
**Time of conclusion of 15-minute infusion.
***1-hour and 4-hour levels were measured only at the beginning of the study.
@Day 1 of aztreonam therapy.
@@Day 3 or 4 of aztreonam therapy.

Values at the beginning and the end of the study were similar, suggesting no significant accumulation of aztreonam given at a dose of 30 mg/kg every 12 hours.

The following pharmacokinetic parameters for aztreonam are listed in the table below: maximal serum concentration (C_{max}), minimum serum concentration (C_{min}), area under the serum concentration-time curve evaluated over one dose interval ($AUC_{0\rightarrow T}$), serum clearance (Cl_s) and serum elimination half-life ($t_{\frac{1}{2}}$).

PHARMACOKINETIC PARAMETERS 30 mg/kg

	C_{max} (µg/ml)		C_{min} (µg/ml)		$t_{\frac{1}{2}}$ (hr)		Cls (ml/min/kg)		AUC_{0-T} (µg.hr/ml)	
	DAY 1	DAY 3/4	DAY 1@	DAY 3/4@@	DAY 1	DAY 3/4	DAY 1	DAY 3/4	DAY 1	DAY 3/4
MEAN	77.5	82.8	23.2	24.3	8.38	6.32	0.75	1.06	499.8	514.9
SEM	5.4	4.8	2.1	3.5	0.66	0.46	0.06	0.06	32.4	36.9
N	26	21	26	21	26	21	26	21	25	21

@Calculated from 8 hour result and elimination half-life.
@@Pre-infusion level.

The mean serum half-life on Day 1 was 8.4 hours. Since aztreonam was given every 12 hours, an increase in serum levels might have occurred on

SQUIBB RESEARCH AND DEVELOPMENT
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TABLE 1

DEMOGRAPHY AND DOSING INFORMATION

SUBJECT NUMBER	SEX	AGE* (DAYS)	WEIGHT (KG)	HEIGHT (CM)	SURFACE AREA (M-SQ)	DOSE** (MG)	DOSE** (MG/KG)	DOSE** (MG/M-SQ)
1	F	2	2.0	46.0	0.161	60.0	30.0	372.7
2	F	1	1.6	43.5	0.139	49.5	30.9	356.1
3	M	1	1.9	45.0	0.155	57.0	30.0	367.7
4	F	2	1.4	39.0	0.126	40.5	28.9	326.6
5	F	1	1.4	38.0	0.123	42.0	30.0	341.5
6	M	4	1.9	42.5	0.151	57.0	30.0	377.5
7	F	1	1.8	43.0	0.148	55.5	30.8	375.0
8	F	1	2.0	44.5	0.159	60.0	30.0	377.4
9	M	1	1.4	39.5	0.125	43.5	31.1	348.0
10	M	1	1.4	37.5	0.122	42.0	30.0	344.3
11	F	3	1.0	37.0	0.102	31.5	31.5	306.8
12	M	2	1.9	45.0	0.155	58.5	30.8	377.4
13	F	2	1.0	37.5	0.102	28.5	28.5	279.4
14	F	1	1.9	44.0	0.154	57.0	30.0	370.1
15	M	1	1.1	41.0	0.111	33.0	30.0	297.3
16	M	4	0.9	35.0	0.094	25.5	28.3	271.3
17	F	1	1.0	35.0	0.099	30.0	30.0	303.0
18	M	1	1.8	46.0	0.152	57.0	31.7	375.0
19	F	4	0.7	33.0	0.080	21.0	30.0	262.5
20	F	1	1.8	46.0	0.154	54.0	30.0	350.6
21	F	2	1.9	43.0	0.152	60.0	31.6	394.7
22	F	1	0.7	32.0	0.079	21.0	30.0	265.8
23	M	1	0.9	35.0	0.094	30.0	33.3	319.1
24	F	2	1.4	40.5	0.126	42.0	30.0	333.3
25	M	1	1.4	41.0	0.127	42.0	30.0	330.7
26	M	1	1.4	41.0	0.127	45.0	32.1	354.3
MEAN		1.7	1.4	40.5	0.128	44.0	30.4	337.7
SEM		0.2	0.1	0.9	0.005	2.6	0.2	7.6
MINIMUM		1	0.7	32.0				
MAXIMUM		4	2.0	46.0				

*: AGE AT FIRST DOSE OF AZTREONAM, STUDY DAY ONE.
**: DOSE WAS INFUSED OVER 15 MINUTES.
DOSING INTERVAL WAS 12 HOURS.

TABLE 2

SERUM AZTREONAM CONCENTRATION RESULTS
(MCG/ML)

SUBJECT NUMBER	TIME (RELATIVE TO THE END OF INFUSION)											
	PRE-INFUSION*		0.0 HOURS		0.5 HOURS		1 HOUR	2 HOURS		4 HOURS	8 HOURS	
	DAY 1	DAY 3/4	DAY 1	DAY 3/4	DAY 1	DAY 3/4	DAY 1	DAY 1	DAY 3/4	DAY 1	DAY 1	DAY 3/4
1												
2												
3												
4												
5												
6												
7												
8												
9												
10												
11												
12												
13												
14												
15												
16												
17												
18												
19												
20												
21												
22												
23												
24												
25												
26												
MEAN	0	24.4	75.5	78.2	66.8	75.0	61.0	54.5	57.3	45.6	32.0	31.5
SEM	0	3.5	5.4	4.4	4.4	4.6	3.5	3.1	3.7	3.3	2.4	3.3

*: 10 MINUTES BEFORE INFUSION.
**: 15 MINUTE INFUSION.
.: THERAPY DISCONTINUED ON STUDY DAY ONE OR TWO.
EXCEPTIONS: SUBJECT 17 AT 4 HOURS, NO SAMPLE COLLECTED
SUBJECT 22 AT 0.5 HOURS END, COLLECTED AT 1 HOUR = 54.8 MCG/ML
SUBJECT 24 AT 0.5 HOURS END, NO VALUE REPORTED

TABLE 5

AZTREONAM PHARMACOKINETIC PARAMETERS

SUBJECT NUMBER	SERUM CONCENTRATIONS (MCG/ML) MAXIMUM DAY 1 (TIME(HRS))	MAXIMUM DAY 3/4 (TIME(HRS))	MINIMUM# DAY 3/4@@	ELIMINATION HALFLIFE (HRS) DAY 1	DAY 3/4	SERUM CLEARANCE (ML/MIN/KG) DAY 1	DAY 3/4
1							
2							
3							
4							
5							
6							
7							
8							
9							
10							
11							
12							
13							
14							
15							
16							
17							
18							
19							
20							
21							
22							
23							
24							
25							
26							
MEAN	78.1+ (0.1)	81.8++ (0.2)	23.2	29.3	6.38	6.32	0.75	1.06
SEM	6.5+ (0.1)	4.5++ (0.1)	2.1	3.5	0.66	0.46	0.06	0.06

#: 12 HOUR DOSING INTERVAL.
@: CALCULATED FROM 6 HOUR RESULT AND ELIMINATION HALFLIFE.
@@: PREINFUSION RESULT.
.: THERAPY DISCONTINUED ON STUDY DAY ONE OR TWO.
+: ONLY CMAX VALUES THAT OCCURRED AT TIME = 0 HOURS WERE USED. N=21.
++: ONLY CMAX VALUES THAT OCCURRED AT TIME = 0 HOURS WERE USED. N=6.

TABLE 6

AZTREONAM PHARMACOKINETIC PARAMETERS

SUBJECT NUMBER	AREA UNDER THE SERUM CONCENTRATION VS TIME CURVE (MCG HR/ML) DAY 1@	Day 1 @@	DAY 3/4@@	VOLUME OF DISTRIBUTION BY AREA (L/KG) DAY 1	DAY 3/4
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					
11					
12					
13					
14					
15					
16					
17					
18					
19					
20					
21					
22					
23					
24					
25					
26					
MEAN	802.5	499.8	518.9	0.488	0.550
SEM	71.0	32.6	36.9	0.027	0.032

#: AUC FROM TIME 0 TO INFINITY.
@@: AUC FROM TIME 0 TO 12 HOURS.
.: THERAPY DISCONTINUED ON STUDY DAY ONE OR TWO.

TABLE 5

AZTREONAM PHARMACOKINETIC PARAMETERS*

SUBJECT NUMBER	AUMC (MCG HR HR/ML)	VD, STEADYSTATE (L/KG)	MRT (HRS) INFUSION@	MRT (HRS) IV BOLUS@
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				
11				
12				
13				
14				
15				
16				
17				
18				
19	---			
20				
21				
22				
23				
24				
25				
26				
MEAN	10842.9	0.478	12.06	11.94
SEM	1669.8	0.026	0.93	0.93

*: DAY ONE OF AZTREONAM THERAPY.
@: 15 MINUTE INFUSION.

TABLE 6

LEG TWO RESULTS, MAINTAINENCE THERAPY
NONCOMPARTMENTAL PHARMACOKINETIC PARAMETER VALUES
VOLUME OF DISTRIBUTION AT STEADYSTATE

SUBJECT	VOLUME OF DISTRIBUTION AT SS (L)	VD SS PER KG BODY WEIGHT (L/KG)	VD SS PER M-SQ OF BSA (L/M-SQ)
1			
2			
3			
4			
5			
6			
7			
8			
9			
10			
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			
24			
25			
26			
MEAN	0.755	0.532	5.871
SEM	0.065	0.034	0.386
N	21	21	21

. - Therapy discontinued on study Day 1 or 2.

TABLE 7

LEG TWO RESULTS, MAINTAINENCE THERAPY
NONCOMPARTMENTAL PHARMACOKINETIC PARAMETER VALUES
MEAN RESIDENCE TIME VALUES

SUBJECT	MEAN RESIDENCE TIME, INFUSION (HR)	CALCULATED MRT, IV BOLUS (HR)
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
11		
12		
13		
14		
15		
16		
17		
18		
19		
20		
21		
22		
23		
24		
25		
26		
MEAN	9.02	8.90
SEM	0.75	0.75
N	21	21

. - Therapy discontinued on study Day 1 or 2.

TABLE 8

DAY 1 URINE AZTREONAM CONCENTRATIONS

PATIENT	TIME (HR)	URINE CONCENTRATION (MCG/ML)
13	NR	
15	NR	
16	NR	
21	NR	
22	NR	
23	NR	
24	NR	
25	NR	
26	NR	
18	0.17	
2	0.50	
19	0.50	
1	1.00	
4	1.00	
5	1.00	
7	1.00	
9	1.00	
10	1.00	
11	1.00	
14	1.00	
3	2.00	
12	2.00	
17	2.00	
20	2.00	
6	3.00	
8	12.00	
MEAN	1.89	253.6
SEM	0.65	22.1
MAXIMUM	12.00	488.7
MINIM	0.17	24.2

NR - Not reported

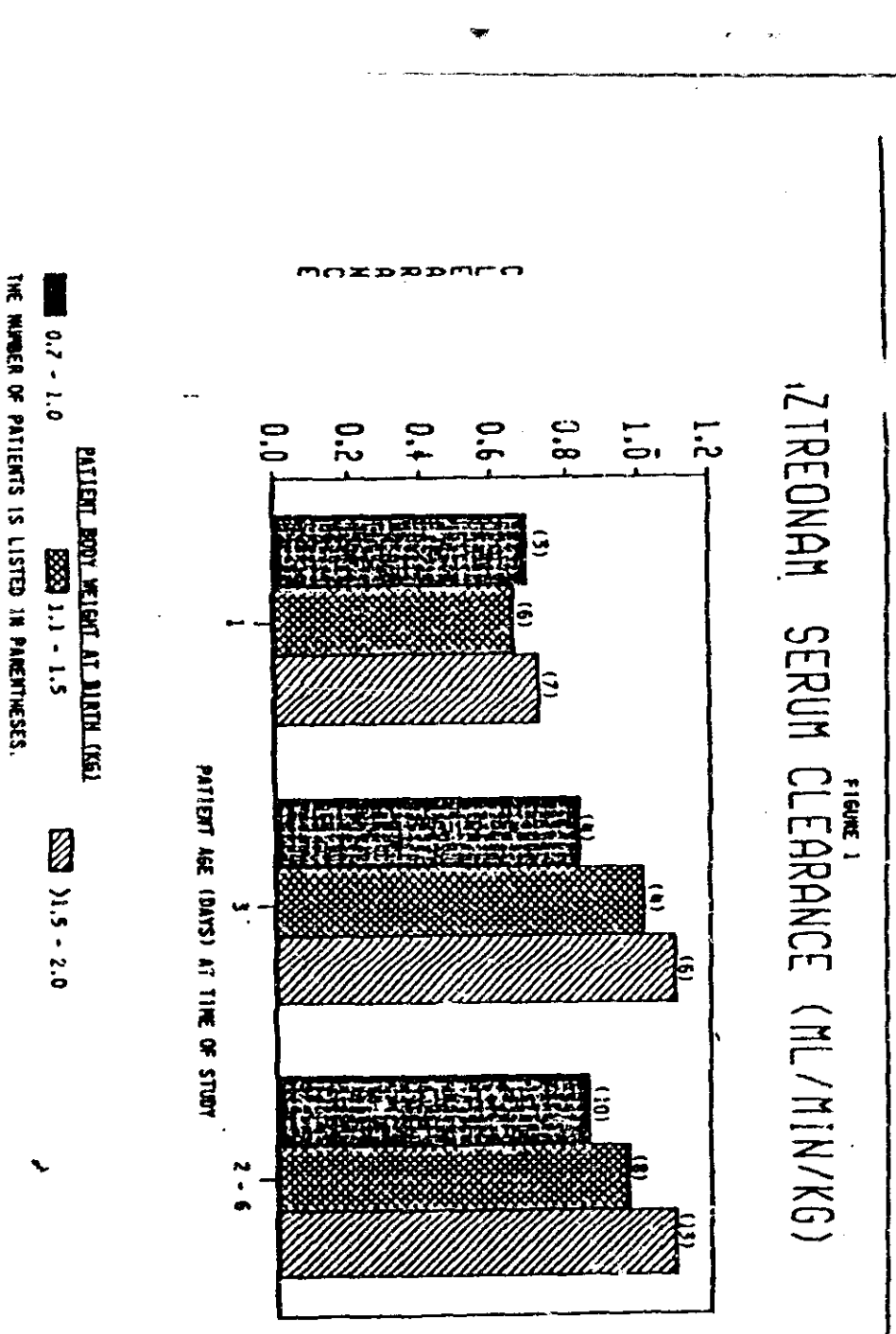
Table 9

RESULTS FOR PATIENTS ONE WEEK OR OLDER AT TIME OF STUDY

AGE AT TIME OF STUDY Patient 11 - 7 Days (Study Day 5) Patient 12 - 8 Days (Study Day 7)		
TIME (HR)	SERUM CONCENTRATION (µg/ml)	
	Patient 11	Patient 12
Pre	24.3	5.9
End of Infusion,0.00	70.4	26.5
0.50	68.4	52.4
2.00	62.6	52.4
8.00	32.7	25.0

	Patient 11	Patient 12
Maximum serum concentration (µg/ml):	70.4	52.4
Time of maximum serum concentration (hr):	0.0	0.5[a]
Serum elimination half-life (hr).	7.06	6.56
Area under serum concentration time curve (hr·µg/ml)·	534.3	412.9
Serum clearance (ml/min/kg):	0.98	1.25
Volume of distribution by area (l/kg):	0.601	0.709
Volume of distribution at steady state (l/kg):	0.564	0.676
Mean residence time, infusion (hr):	9.68	9.14
Mean residence time, bolus (hr):	9 56	9.02

[a]The concentration at 0.5 and 2.0 hours were identical, probably because the infusion was not completed at the scheduled time.

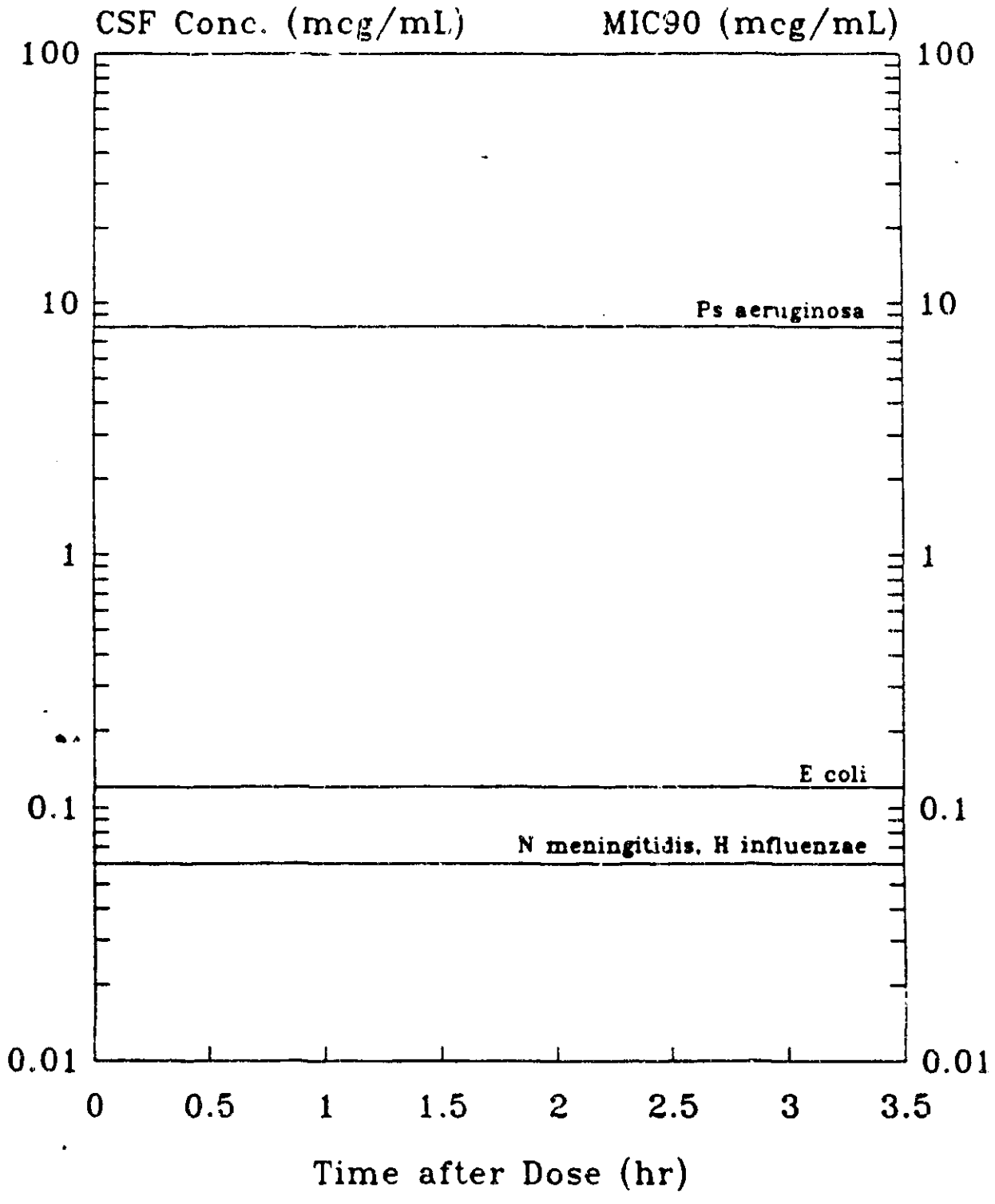


Protocol 18554-58 Addendum D

APPENDIX X

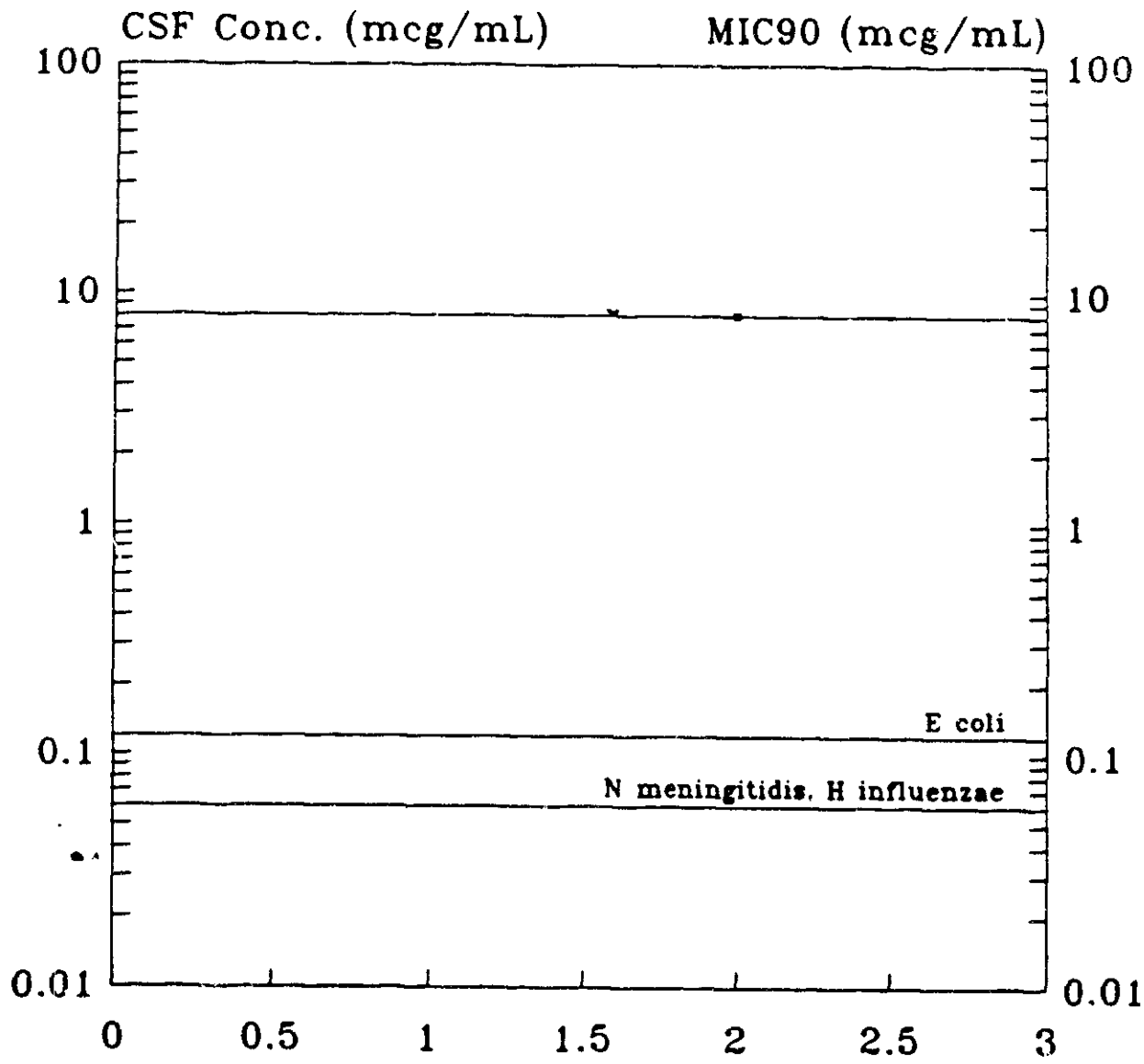
Protocol 18,554-32

CSF Concentrations in Pediatric Patients
Inflamed Meninges, Single 30mg/kg IV Dose



Protocol 18,554-588

CSF Concentrations in Pediatric Patients with Inflamed Meninges



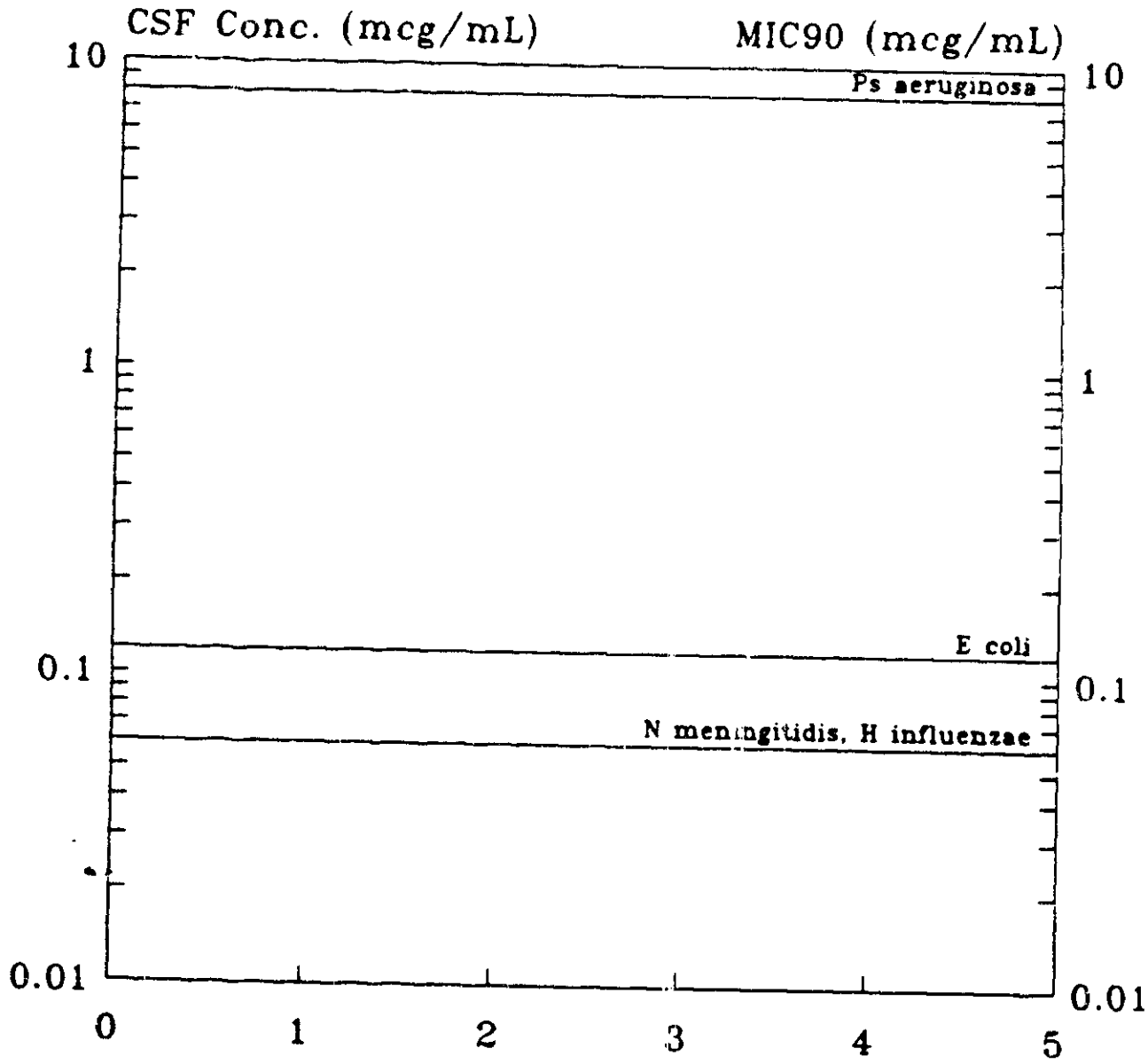
• Day 2 × Day 3 ◇ Day 4

△ Day 5 + Day 7

IV Doses Ranged from 25–50 mg/kg Daily

Protocol 18,554-16A Addendum B

CSF Concentrations in Adult Patients
Inflamed Meninges

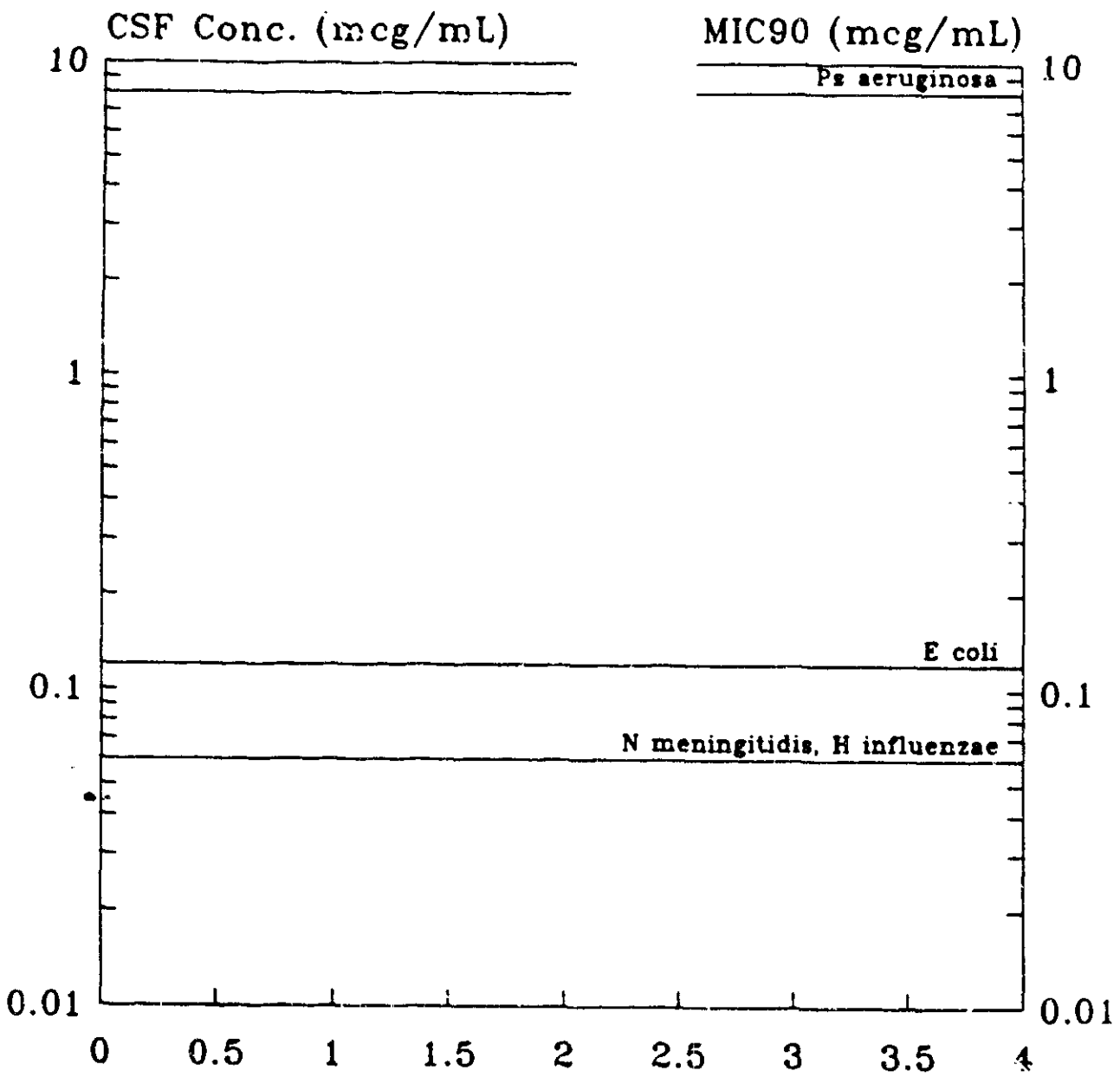


• Pat. 7709-017 ◇ Pat. 7516-001

Pat. 7709-017: 1 g q6h IV; 1 hr sample, day 1; 4 hr sample, day 15 of therapy
Pat. 7516-001: 2 g q6h IV; day unknown

Protocol 18,554-52

CSF Concentrations in Pediatric Patients
Inflamed Meninges, Single IV Dose



Time after Beginning of IV Infusion(hr)

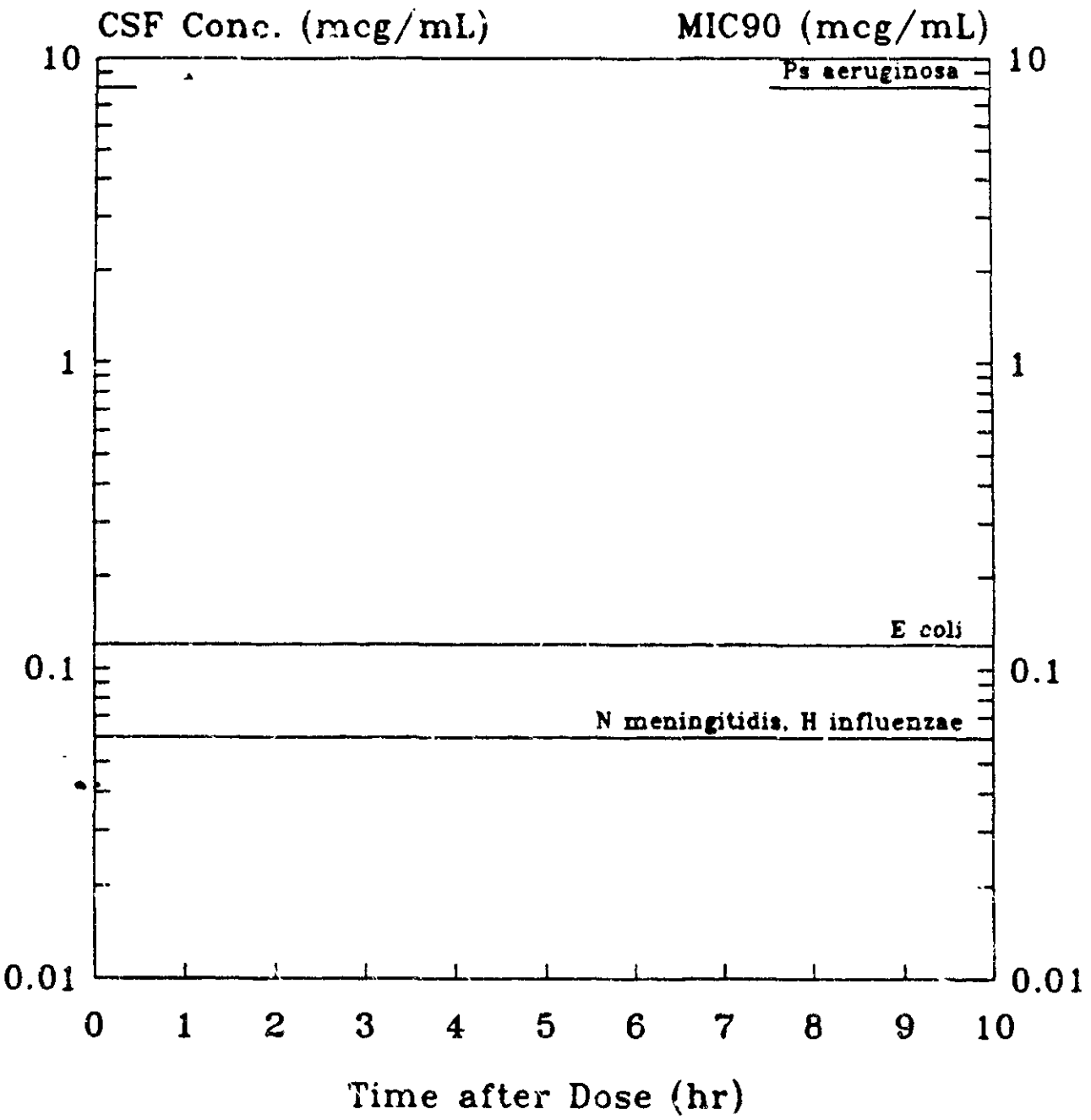
• 30 mg/kg ✡ 50 mg/kg

Dose infused over 30 minutes

N 50580 Bio -2

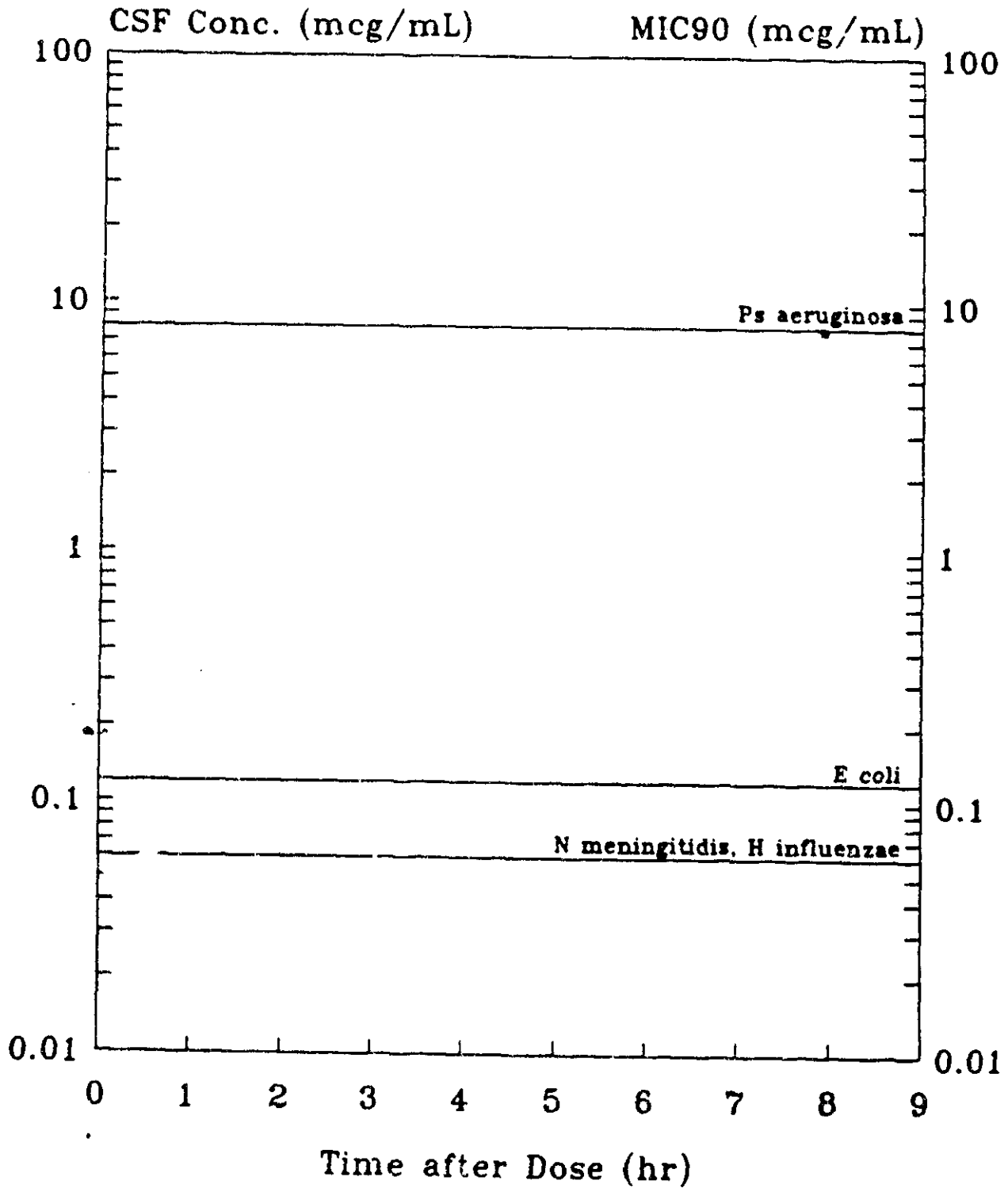
Protocol 18,544-29

CSF Concentrations in Adult Patients
Single 2 g IV Dose



Protocol 18,554-51

CSF Concentrations in Adult Patients
Inflamed Meninges, Single 2 g IV Dose



11 Pages

Purged

M E M O R A N D U M

DEPARTMENT OF HEALTH & HUMAN SERVICES
Public Health Service
Food and Drug Administration
Center for Drugs and Biologics
Office of Drug Standards

[redacted]

DATE : FEB 10 1986

TO : Edward Tabor, M.D.
Director,
Division of Anti-Infective Drug Products
(HFN-815)

FROM : Jerome P. Skelly, Ph.D.
Director,
Division of Biopharmaceutics
(HFN-220)

SUBJECT: Biopharmaceutics Recommendation of Approval;
Aztreonam (AZACTAM); NDA 50-580

I. BACKGROUND

Aztreonam is the first member of a new class of antibiotics classified as monobactams. It is a totally synthetic bacterial antibiotic with activity against a wide spectrum of gram-negative aerobic pathogens.

AZACTAM for Injection is a soluble sterile nonpyrogenic white powder containing approximately 780 mg L-arginine per gram of aztreonam for intramuscular or intravenous use following constitution.

Depending upon the type and severity of infection in the patient with a normal renal function, the following aztreonam dosage is recommended:

A) 0.5 g or 1 g b.i.d. or t.i.d. (urinary tract infection)

B) 1 g or 2 g b.i.d. or t.i.d. (moderately severe systemic infections)

C) 2 g t.i.d. or q.i.d. (severe systemic or life-threatening infections)

The maximum recommended dose is 8 g per day with single doses greater than 1 g being recommended for IV administration.

II. STUDIES

The sponsor in NDA 50-580 filed a total of 34 bioavailability/pharmacokinetic studies. Seven of those studies were classified as pivotal while 27 were classified as supportive. The reviewed studies have been further categorized and are summarized as follows.

A) Dose Proportionality (Single Dose) Studies

Three dose proportionality studies were conducted in normal healthy volunteers. The dosage ranges that were studied adequately covered the individual IM or IV doses within the package insert's proposed b.i.d., t.i.d., or q.i.d dosing regimens.

Specifically, one study (parallel study design; n = 6 per dose level) compared [redacted] 0.5, 1, 2, and [redacted]g aztreonam given as 3-minute single dose IV infusions. Another study (crossover study design; n = 6) compared [redacted] 0.5 and 1 g aztreonam given as 30 minute single dose IV infusions. A third study (parallel study design; n = 6 per dose level) compared [redacted], 0.5 and 1 g aztreonam given as single dose IM injections. These studies provided aztreonam single dose pharmacokinetic data (e.g. t1/2 = 1.5-2 hrs, Tmax = 1 hr, etc.) and demonstrated the drug to be dose proportional over the dosage ranges and the two routes of administration studied.

B) Metabolism and Excretion/Pharmacokinetic Study

A crossover study was conducted in six healthy male volunteers where single doses of ^{14}C-aztreonam (0.5 g) were administered as an IV 2-minute infusion and as an IM injection. This study demonstrated that aztreonam is 100% bioavailable by the IM route and that approximately 90% or more of the administered ^{14}C labeled dose can be accounted for (in urine (total = 77%, parent = 67%) and feces (total = 13%, parent = 1%)). The t1/2 of the major metabolite was determined to be 25 hours (i.e. bio-inactive, open beta-lactam ring hydrolysis product).

C) Multiple Dose Pharmacokinetic Studies

Two separate multiple dose studies were conducted in normal healthy volunteers where drug was administered for seven days (22 doses). One study (parallel study design; n = 9 per dose regimen) assessed aztreonam's pharmacokinetics following IM dosing schedules of 0.5 and 1 g t.i.d. The other study (parallel study design; n = 9 per dose regimen) assessed the drug's pharmacokinetics following 0.5 and 1 g t.i.d. dosing regimens where aztreonam was given as 2-minute IV infusions. These studies demonstrated there was no aztreonam accumulation using the studied dosing regimens. Some accumulation did occur for the major metabolite. Drug serum protein binding was shown to be 56%.

D) Special Studies

Information obtained from these studies was incorporated in the proposed package insert.

1) In Disease States Studies

Single dose and multiple dose IV studies where aztreonam was administered to patients (including elderly) with different degrees of renal dysfunction, showed that renal impairment significantly effected the elimination of aztreonam warranting drug dose adjustment based upon renal function. Provided also were studies that assessed the effects of hemodialysis, peritoneal dialysis, hepatic disease, and drug administered to patients undergoing cancer therapy.

Hemodialysis and peritoneal dialysis studies demonstrated the amounts of drug that could be eliminated by these prccesses. The study in hepatic disease showed an increase in the elimination half-life of the drug to about 3.2 hours in alcoholic patients as compared to 1.9 hours in healthy subjects. Studies in cancer patients with normal renal function demonstrated that aztreonam was handled as similarly in normal healthy volunteers.

2) Drug Interaction Studies

Drug interaction studies in healthy volunteers were conducted where aztreonam was intravenously administered with and without probenecid, furosemide, gentamicin, clindamycin, metronidazole, nafcillin, and cephradine. None of these interaction studies demonstrated significant effects on the overall pharmacokinetics of aztreonam or vice versa.

III. Renal Impaired Patients

Serum levels and the pharmacokinetics of aztreonam and the major metabolite have been determined in renally impaired patients. Evaluation of such study results from a pharmacokinetic perspective (see review Sections II.G, III. 4a-4d, 5a-5c, 7a-7b, & 8a-8b, IV and V.E) suggest that if the proposed package insert dosing recommendations for renally impaired patients are clinically acceptable, then the labeling should be updated to include the following (or similar text) in the Renal Impairment Section.

"Studies in renally impaired patients have demonstrated that high trough drug levels can occur along with some potential for accumulation of aztreonam's open beta-lactam ring metabolite. In patients with severe renal dysfunction it is therefore prudent to monitor aztreonam and its metabolite before increasing doses."

III. OVERALL RECOMMENDATION

The Division of Biopharmaceutics (DB) finds NDA 50-580 approvable in that it meets the Agency's Bioavailability and Bioequivalence Requirements cited under 21 CFR 320. However, DB recommends, based upon a pharmacokinetic perspective, that additional information addressing dose adjustment in renal impairment, as indicated above, is warranted. This as well as the other final printed labeling issues (review Section V) should be brought the attention of the reviewing medical officer.

Jerome P. Skelly, Ph.D.
Director,
Division of Biopharmaceutics

Prepared by John P. Hunt
Initialed by C.T. Viswanathan, Ph.D. CTV 6/5/86

cc: HFN-520 (Dr. Skelly), HFN-225 (Hunt), Drug File, Review File, Chron

JPH:smj:[redacted]:6/5/86

Aztreonam Injection
(AZACTAM)
NDA 50-580
Reviewer: John P. Hunt

[redacted]

E.R. Squibb & Sons, Inc.
Princeton, NJ 08540
Submission Dated:
August 27, 1984

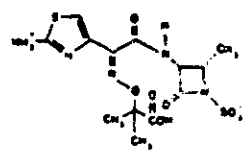
Review of Pharmacokinetic/Bioavailability Studies & Labeling

I. Background:

AZACTAM (aztreonam, SQ26,776) is the first member of a new class of antibiotics classified as monobactams. These agents were originally isolated from Chromobacterium violaceum. AZACTAM is a totally synthetic bactericidal antibotic with activity against a wide spectrum of gram-negative aerobic pathogens.

The monobactams, having a unique monocyclic beta-lactam nucleus, are structurally different from other beta-lactam antibiotics (e.g., penicillins, cephalosporins, cephamycins). The sulfonic acid substituent in the 1-position of the ring activates the beta-lactam moiety; an aminothiazolyl oxime side chain in the 3-position and a methyl group in the 4-position confer the specific antibacterial spectrum and beta-lactamase stability.

Aztreonam is designated chemically as (z)-2-[[[(2-amino-4-thiazolyl)[[(2S, 3S)-2-methyl-4-oxo-1-sulfo-3-azetidinyl]carbamoyl]methylene]amino]oxy]-2-methyl-propionic acid. Structural formula:



$C_{13}H_{17}N_5O_8S_2$ MW 435.42 CAS-78110-38-0

AZACTAM for Injection (Aztreonam for Injection) is a sterile nonpyrogenic white powder, containing approximately 780 mg L-arginine per gram of aztreonam, for intramuscular or intravenous use following constitution. The powder is sodium-free. Aqueous solutions of aztreonam have a pH in the range of 4.5 to 7.5.

Depending upon the type and severity of infection being treated, doses of aztreonam can be given to adult patients (with normal renal function) that range between i) 0.5 g or 1 g b.i.d. or t.i.d.(urinary tract infection), ii) 1 g or 2 g b.i.d. or t.i.d. (moderately severe systemic infections) and, iii) 2 g t.i.d. or q.i.d. (severe systemic or life threatening infections). The maximum recommended dose is to be 8 g per day. Single doses greater than 1000 mg are recommended to be given by the IV route.

In this Division of Biopharmaceutics (DB) review, studies that were identified by the sponsor as being pivotal pharmacokinetic (PK) studies (see Appendix A) have been critically evaluated. Also reviewed in this document, are the sponsor's other supportive PK studies (Appendix A) from which some information has been obtained to support labeling claims that are in the proposed package insert (Appendix B). Generally for the supportive PK studies covered in this document, only the sponsor's submitted study summarizes and appropriate tables are provided. The supportive studies' summary results have been compared to the results of the pivotal PK studies, where appropriate, and comments are generally only made if there are observed differences or decrepancies.

NOTE: Appendix B has only those portions of the sponsor's proposed package insert (filed 5/13/86) that have been reviewed by DB. Comments recommending the inclusion of additional information or modification of existing proposed labeling claims are given in Section V of this review.

Table 1. Pharmacokinetic Analysis of Aztreonam Data

mean (S.E.M.)

Parameter	Biliary Cirrhosis	Alcoholic Cirrhosis	Normal Subjects
C_{max}, ug/ml	103.20 ± 13.51	115.40 ± 16.43	114.40 ± 14.49
$AUC_{0-12\ hr}$ ugxhr/ml	237.60 ± 21.16	231.10 ± 21.10	189.40 ± 15.00
Distribution			
*Extent			
V_1, liters/kg	0.12 ± 0.02	0.08 ± 0.02	0.06 ± 0.02
V_{DSS}, liters/kg	0.18 ± 0.02	0.18 ± 0.02	0.13 ± 0.02
$V_{D\ AREA}$, liters/kg	0.19 ± 0.02	0.22 ± 0.03	0.17 ± 0.01
serum protein binding %	69.62 ± 1.36	69.13 ± 1.72	73.02 ± 2.02
*Rate			
$t_{\frac{1}{2}\alpha}$, hr^{-1}	0.28 ± 0.09	0.36 ± 0.17	0.14 ± 0.04
k_{12}, hr^{-1}	1.82 ± 0.72	3.21 ± 1.31	3.61 ± 1.01
k_{21}, hr^{-1}	2.16 ± 0.53	1.12 ± 0.27	1.87 ± 0.27
Elimination			
*Extent			
12-hr urinary excr. % of Dose	54.41 ± 6.73	**75.53 ± 7.22	62.41 ± 5.55
serum clearance, ml/min/kg	1.00 ± 0.08	***0.82 ± 0.04	1.08 ± 0.12
renal clearance, ml/min/kg	0.55 ± 0.10	0.63 ± 0.08	0.69 ± 0.11
nonrenal clearance, ml/min/kg	**0.45 ± 0.07	0.19 ± 0.05	**0.39 ± 0.01
*Rate			
$t_{\frac{1}{2}\beta}$, hr_{-1}	***2.17 ± 0.06	***3.24 ± 0.56	1.89 ± 0.17
k_{10}, hr^{-1}	0.61 ± 0.10	1.47 ± 0.78	2.10 ± 1.13

*Significantly different from biliary cirrhosis mean (P<0.05).
**Significantly different from alcoholic cirrhosis mean (P<0.01 and P<0.05 for biliary cirrhosis and healthy subjects respectively).
***Significantly different from the mean for healthy subjects (P<0.05).

TABLE 6

MEAN CUMULATIVE URINARY EXCRETION OF SQ 26,992
as Measured by HPLC
(in mg)

		Biliary	Alcoholic	Normal
0 to 12 Hrs	Mean:	14.482	15.760	19.028
	SEM:	(2.4036)	(1.5640)	(3.3327)
	n:	5	6	6
	% Dose	1.5%	1.6%	1.9%

Figure 1
Effect of Hepatic Disease on Serum Pharmacokinetics of 3-Minute Intravenous Infusions of 1 gm Aztreonam

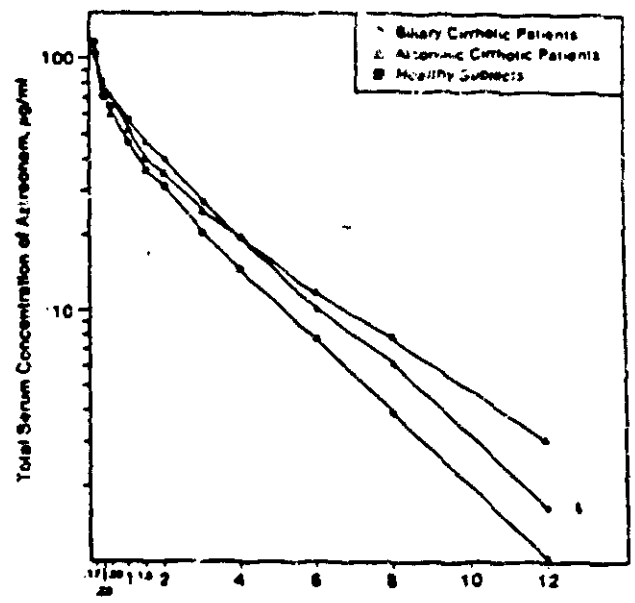
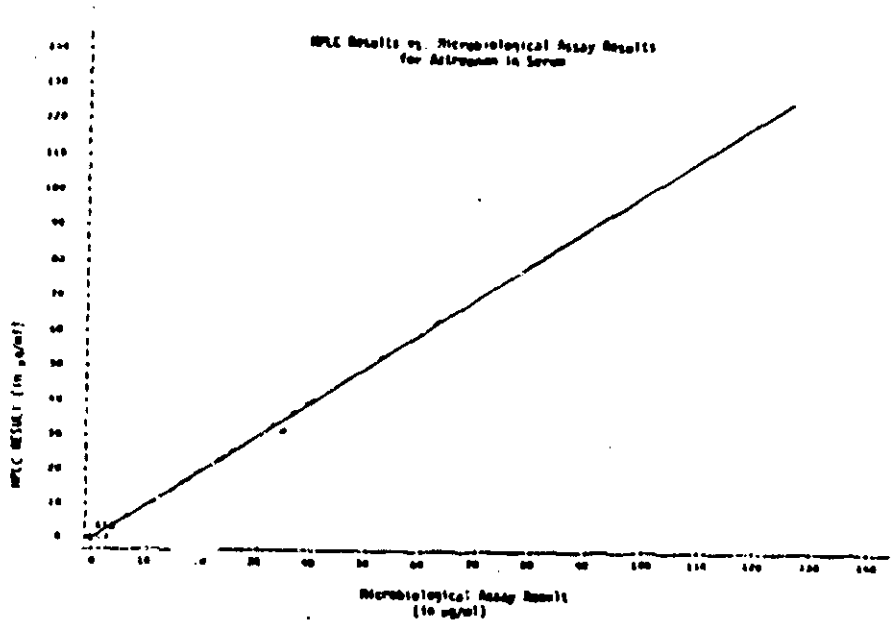


TABLE 1A

SUMMARY OF
SERUM CONCENTRATION OF AZTREONAM
As measured by Microbiological Assay
(Means and S.E.M.'s)
(in ug/ml)

Time After Infusion (in hours)	Biliary Cirrhosis Patients n= 6	Alcoholic Cirrhosis Patients 6	Healthy Subjects 6
Prior to Infusion	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
0.17	103.2 (13.5)	109.8 (17.7)	114.4 (14.5)
0.33	82.5 (11.0)	77.0 (9.6)	70.9 (2.8)
0.50	72.2 (7.2)	60.1 (4.6)	64.1 (2.9)
1.00	57.5 (5.5)	55.1 (7.1)	46.1 (3.3)
1.50	46.7 (3.7)	40.0 (3.2)	36.7 (2.2)
2.0	40.7 (3.1)	36.0 (3.1)	31.2 (2.5)
3.0	27.3 (2.4)	25.2 (2.3)	20.6 (1.8)
4.0	19.9 (1.7)	19.6 (2.2)	14.8 (1.7)
6.0	10.7 (1.1)	11.6 (1.4)	7.6 (1.4)
8.0	6.1 (0.6)	7.6 (1.0)	3.9 (0.9)



Solid line represents HPLC = Micro.

"A" represents 1 observation, "B" represents 2 observations, etc.

VII. ADVERSE REACTIONS

Four of the 18 volunteers experienced adverse reactions during the study. Patient 1 (a biliary cirrhotic patient) had moderate abdominal pain 1 hour after eating on the same day aztreonam was administered. The pain subsided within 30 minutes without treatment and was considered by the investigator to be related to the patients' cholecystectomy. Subject 13 (a healthy subject) had mild abdominal discomfort after dinner 10 hours following aztreonam administration. The discomfort lasted 1 hour and disappeared without treatment. This was also not considered to be related to aztreonam by the investigator. Subject 15 (a healthy subject) experienced mild fatigue and mild difficulty in concentration on Day 3. Both of these effects were considered to be work related. Subject 17 (a healthy subject) had several loose stools for 4 hours beginning 14 hours after drug infusion. The condition subsided

Attachment I

Mezlocillin kinetics are altered in hepatic disease (Bunke et al. 1983). In patients with alcoholic cirrhosis, the terminal half-life of mezlocillin was almost three times longer than that in healthy subjects and nonrenal clearance was markedly reduced (by 90%). The authors recommended dosage reduction of mezlocillin in hepatic patients according to the following equation:

$$F_p = \frac{AUC_N}{AUC_p}$$

where F_p is the dose fraction of a drug for a given patient with decreased clearance of that drug. AUC_p is the AUC for that patient and AUC_N is the AUC for patients with normal clearance of the drug. The dose fraction in that study for patients with hepatic disease was 0.51. The dose fraction is multiplied by the normal dose to obtain the reduced dose for cirrhotic patients. Thus the patients in that study would receive only half the usual dose. This method of calculating dosage reduction assures that the AUC in patients with reduced clearance will remain constant and equal to that in normal patients. If, on the basis of clinical status or anticipated duration of therapy, dosage reduction of aztreonam becomes desirable, dosage could be reduced according to the following formula (AUCs from Table 6):

$$F_p = \frac{AUC_N}{AUC_p} = \frac{189}{231} = 0.82$$

Thus, for alcoholic cirrhotics, the dose would be reduced by 18%.

Another method for calculating dosage reduction is based on comparison of serum clearances, i.e., the dose fraction is derived by dividing the serum clearance in patients by the serum clearance in normals (Aronoff et al. 1981). For the present study (serum clearances from Table 8) the dose fraction becomes: 0.82 ÷ 1.08 = 0.76, and the dose for alcoholic cirrhotics would be 76% of the normal dose. In practice, the physician, who was concerned about the dose of aztreonam in an alcoholic cirrhotic patient, could reduce the dose by 20-25% and be reasonably sure that the AUC in that patient would be similar to that in patients with normal clearances.

DEPARTMENT
Department of Clinical Pharmacology

SECTION
Division of Medical Affairs

DATE OF WRITING REPORT
April 1, 1983

INVESTIGATIVE
RMB-860

NUMBER ASSIGNED BY SPONSOR
SQ 26,776 (Aztreonam)

TITLE
Report on Comparison of Safety, Pharmacokinetics, and Serum and Urine Bactericidal Activity of Intravenous Aztreonam and Moxalactam in Healthy Subjects. Study Protocol # 18554-23

AUTHORS
Edward A. Swabb, M.D., Ph.D., May Frantz, Ph.B., and Tricia Yen, M.S.

INVESTIGATORS, M.D., Department of Medicine, University of Zurich

ABSTRACT
Aztreonam and moxalactam were each administered as a single 2000-mg intravenous infusion over 30 minutes to 6 healthy male volunteers according to a two-way crossover study design with a 7-day washout period between drug treatments. To assess the safety of the drug treatments, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and at frequent intervals after each drug treatment.

Aztreonam and moxalactam were tolerated well by 6 healthy male subjects. Possible drug-related adverse reactions after administration of aztreonam consisted of mild diaphoresis (1 subject) and mild fatigue (2 subjects), and after administration of moxalactam consisted of mild diarrhea and dizziness (1 subject) and mild diarrhea and flatulence (1 subject). These findings were reversible without specific treatment.

The pharmacokinetic profiles of aztreonam and moxalactam were assessed by measuring aztreonam and moxalactam (sum of R and S epimers) concentrations in multiple samples of serum and urine after administration of the antibiotics. Assays were performed by the clinical investigator using a high-pressure liquid chromatography method. Mean values for the concentrations of aztreonam and moxalactam in serum and urine are shown in Table I.

Table I

Time After Start of Infusion, hr	Serum[c] Aztreonam, ug/ml	Serum[c] Moxalactam, ug/ml
Pre	0 ± 0	0 ± 0
0.5[a]	137.1 ± 2.3	169.2 ± 3.3
2	51.8 ± 1.7	63.0 ± 1.7
6	13.3 ± 0.6	18.0 ± 0.9
8	6.7 ± 0.2	8.6 ± 0.2
12	1.8 ± 0.2	2.2 ± 0.1
Time After Start of Infusion, hr	**Urine[c] Aztreonam, ug/ml**	**Urine[c] Moxalactam, ug/ml**
Pre	0 ± 0	0 ± 0
0-0.5[b]	6120 ± 1367	8534 ± 1103
0.5-2	6010 ± 1349	9479 ± 1652
2-6	2139 ± 573	2977 ± 754
6-8	957 ± 278	1475 ± 121
8-12	311 ± 43	735 ± 120

[a] At the end of the 30-minute infusion.

[b] Collected during the 30-minute infusion.

[c] Values are arithmetic mean ± SEM for 6 subjects.

Maximum serum concentrations (C_{max}), areas under the serum concentration-time curve (AUC), elimination half-life ($t_{1/2}$), and urinary recovery are shown in Table II. Although moxalactam gave statistically significantly greater mean values for C_{max} and AUC, none of the differences shown in Table II was considered to be of therapeutic importance.

Table II

Parameter[a]	Aztreonam	Moxalactam	p[b]
C_{max}, ug/ml	137.1 ± 2.3	169.2 ± 3.3	<0.01
AUC_{0-12hr}, ug × hr/ml	343.1 ± 5.5	426.8 ± 7.1	<0.01
$t_{1/2}$, hr	2.07 ± 0.11	2.01 ± 0.07	NS
Urinary recovery, % of dose, 0-8 hr	55.5 ± 5.4	64.8 ± 4.3	NS

[a] Values are arithmetic mean ± SEM for 6 subjects.

[b] Based upon analysis of variance for the crossover design.

The pharmacokinetics of aztreonam described in this study were consistent with previously reported results for 30-minute intravenous infusions of a 2-gram dose in healthy volunteers (Protocol 18554-18).

Serum and urinary bactericidal titers were determined by the clinical investigator at the same times as shown in Table I for the six test organisms shown in Table III.

Table III

Bacterial Strain	Aztreonam MIC, ug/ml	Aztreonam MBC, ug/ml	Moxalactam MIC, ug/ml	Moxalactam MBC, ug/ml
Escherichia coli	0.06	0.125	0.125	0.25
Klebsiella pneumoniae	0.06	0.125	0.125	0.5
Proteus mirabilis	0.008	0.016	0.125	0.125
Serratia marcescens	0.06	0.125	0.125	0.5
Pseudomonas aeruginosa	8	16	16	32
Enterobacter cloacae	16	32	16	32

Table IV

Time, hr	Aztreonam *E. coli*	*Kleb. pneum.*	*Proteus mirabilis*	*Serratia marcescens*	*Pseudo. aeruginosa*	*Ent. cloacae*
Pre	<2	<2	<2	<2	<2	<2
0.5	144	45	57	57	6	4
2	72	18	28	32	2	2
6	16	10	7	57	<2	2
8	11	7	4	16	<2	<2
12	6	4	3	7	<2	<2
	Moxalactam					
Pre	<2	<2	<2	<2	<2	<2
0.5	45	40	32	72	2	9
2	28	23	20	45	<2	6
6	11	6	7	18	<2	2
8	8	3	6	8	<2	2
12	6	<2	2	2	<2	<2

Table V

Time, hr	Aztreonam *E. coli*	*Kleb. pneum.*	*Proteus mirabilis*	*Serratia marcescens*	*Pseudo. aeruginosa*	*Ent. cloacae*
Pre	<2	<2	<2	<2	<2	<2
0-0.5	2048	1552	1552	675	194	64
0.5-2	2048	1625	2299	813	456	102
2-6	1149	813	575	406	128	40
6-8	456	256	362	161	64	7
8-12	322	203	144	72	28	3
	Moxalactam					
Pre	<2	<2	<2	<2	<2	<2
0-0.5	9195	2580	362	228	28	81
0.5-2	12321	6502	322	256	32	203
2-6	5161	2048	181	144	23	72
6-8	3251	1625	102	102	11	36
8-12	1149	512	64	64	4	18

The bactericidal activity of aztreonam in humans (Tables IV and V) supports, in patients with normal renal function, a 2-gram q12h intravenous dosage regimen for systemic infections due to *E. coli*, *K. pneumoniae*, *P. mirabilis*, and *S. marcescens* having MIC's of 0.06, 0.06, 0.06 and 0.06 ug/ml, respectively. Therapy of systemic infections due to the test strains of *P. aeruginosa* (MIC = 8 ug/ml) and *E. cloacae* (MIC = 16 ug/ml) would appear to require more frequent administration and perhaps higher doses of aztreonam, in patients with normal renal function. Uncomplicated urinary infections by the test organisms might be treatable with aztreonam using a 0.5-gram q12h intravenous dosage regimen, in patients with normal renal function. However, these suggestions should be considered tentative, pending results of ongoing therapeutic trials in patients.

Aztreonam and moxalactam, administered as single, 2000-mg intravenous doses to healthy male subjects, had similar safety, pharmacokinetic, and bactericidal activity profiles in the present study. However, comparison of safety and efficacy of these two compounds in infected patients awaits results of ongoing clinical trials.

Appendix A

Study No.	Study Type	NDA Vol.	NDA Report First Page	Comments
1) 18,554-1	Pivotal			Study for meeting CFR 320 Bio-Regs (Single dose IV bolus dose proporationality study)
2) 18,554-2	Pivotal			Study for meeting CFR 320 Bio-Regs (Metabolism/excretion; IM vs IV bio-study)
3) 18,554-3	Pivotal			Study for meeting CFR 320 Bio-Regs (Single dose IM dose proportionality study)
4) 18,554-4	Pivotal			Study for meeting CFR 320 Bio-Regs (IV bolus multiple dose study)
5) 18,554-5	Pivotal			Study for meeting CFR 320 Bio-Regs (IM multiple dose study)
6) 18,554-18	Supportive			Single dose IV infusion dose proportionality study
7) 18,554-8	Pivotal			Single dose IV renal impairment study
8) 18,554-9	Pivotal			Single dose IV hepatic disease study
9) 18,554-23	Supportive			PK IV infusion comparison study with moxolactam
10) 18,554-7	Informativ			Oral vs IV bio-study
11) 18,554-68	Supportive			Study in healthy elderly.
12) 18,554-38	Supportive			IV multiple dose study in elderly patients with renal impairment
13) 18,554-24	Supportive			Single dose IV renal impairment study
14) 18,554-25	Supportive			Hemodialysis/peritoneal dialysis study
15) 18,554-11/A and 14/B	Supportive			Renal impairment study

16)	18,554-27/A and 31/A	Supportive	Renal impairment study
17)	18,554-6	Supportive	Drug interaction study-probenecid
18)	18,554-19	Supportive	Drug interaction study-flurosemide
19)	18,554-46	Supportive	Drug interaction study-gentamicin
20)	18,554-47	Supportive	Drug interaction study-clindamycin
21)	18,554-48	Supportive	Drug interaction study-metronidazole
22)	18,554-49	Supportive	Drug interaction study-nafcillin
23)	18,554-59	Supportive	Drug interaction study-cephradine
24)	18,554-29	Supportive	CSF study
25)	18,554-51	Supportive	CSF study
26)	18,554-12	Supportive	Biliary excretion study
27)	18,554-54	Supportive	Bronchial secretion study
28)	18,554-33	Supportive	Excretion in human milk
29)	18,554-34	Supportive	Amniotic fluid, fetal serum and placenta study
30)	18,554-39	Supportive	Blister fluid study
31)	18,554-25/A	Supportive	Prostrate, urinary bladder tissue level study
32)	18,554-20/A	Supportive	Cancer patient multiple dose study
33)	?	Supportive	Human bone and synovial fluid level study
34)	?	Supportive	Human kidney tissue level study

6 Pages

Purged

II. Pivotal Pharmacokinetic Studies:

A. Study Protocol #18,554-1 (Pivotal Study)

1. Title: Ascending dose intravenous safety and pharmacokinetic study of aztreonam healthy subjects.

2. Objective: To define aztreonam's pharmacokinetics, dose proportionality, urinary excretion and safety following increasing intravenous doses.

3. Study Design: Aztreonam was administered as 3-minute intravenous infusions to 36 healthy male volunteers (six groups of 6 subjects each) as single doses of [redacted]500, 1000, 2000 and [redacted] mg. Each group received a separate dose of aztreonam. Drug was supplied as a sterile powder blend of aztreonam and L-arginine (weight ratio = 1.0/0.78) for reconstitution in sterile water. The volume injected for each dose was 10 cc except for the [redacted] mg dose where 13 cc were given. Twelve (12) additional subjects received matching saline placebo injections. To assess the safety of aztreonam, physical and electrocardiographic examinations, injection site evaluations, monitoring of vital signs, and clinical laboratory tests were conducted before and at frequent intervals after each dose of the drug. Approximately 250 ml of water was ingested at the time of drug administration and at 1 and 2 hours post-dose to promote urine formation.

The 36 male subjects receiving aztreonam in this study ranged in age from [redacted] years (mean, 22 years), and their body weights ranged from 55.0 to 92.5 kg (mean, 73.4 kg). The 12 male subjects receiving placebo ranged in age from 18 to 25 years (mean, 21 years), and had body weights ranging from 66.6 to 95.8 kg (mean, 79.9 kg).

Serum samples were serially collected over 24 hours from an antecubital vein which was contralateral to the arm where the drug was injected. Urine samples were collected over 72 hours. Both serum and urine samples were analyzed by a microbiological agar diffusion assay method and a HPLC assay method (see Attachment I). The microbiological assay for this study had a lower quantitation limit of [redacted] mcg/ml in serum and [redacted] mcg/ml in urine. The quantitation limits for the HPLC method were [redacted] mcg/ml and [redacted] mcg/ml, respectively.

Pharmacokinetic (PK) parameters were determined using a two-compartment open model. Specifically, PK analyses were carried out as follows:

Because SQ 26,776 was infused over 3 minutes, the exponential equation, $C = Re^{\alpha t} + Se^{\beta t}$, was used to express the serum concentration (C, ug/ml) vs. time (t, hr) data. Curve stripping of C vs. t data and optimization of the coefficients R, S, α and β by nonlinear regression analysis were performed using the computer

programs AUTOAN and NONLIN (Metzler, Elfring and McEwen, 1974) on an IBM 370 digital computer. All serum data were weighted according to their reciprocals for computer analysis. The relationships between the coefficients R and S and the coefficients A and B for this model were: $A = (R\alpha T)/(1-\exp(-\alpha T))$, $B = (S\beta T)/(1-\exp(-\beta T))$ where α and β were hybrid rate constants and T was the duration of the infusion. Volume of distribution of the central compartment was expressed as V_1 = Dose/((A+B)W), where W was body weight in kg. Volume of distribution based on area under the serum concentration-time curve was expressed as Dose/(βW(A/α+B/β)). Volume of distribution at steady-state was expressed as $V_{ss}=V_1(1+(K_{12}/K_{21}))$, where the intercompartmental rate constants where expressed as $K_{21}=(A\beta+B\alpha)/(A+B)$ and $K_{12}=\alpha+\beta-K_{21}-K_{10}$. In addition, the rate constant for elimination from the central compartment, was expressed as $K_{10}=\alpha\beta/K_{21}$. Half-lives for the distribution and elimination phases of the serum concentration-time data were expressed as $t_{1/2\alpha}=(\ln 2)/\alpha$ and $t_{1/2\beta}=(\ln 2)/\beta$, respectively. Serum clearance was expressed as $CL=V_1K_{10}/W$. The area under the serum concentration-time curve, AUC, was calculated using the trapezoidal rule with C=R+S at t=0.

Pharmacokinetic data analyses for the other intravenous studies that were submitted were analyzed similarly.

Clinical portions of the study were conducted by Arthur Sugerman, M.D., The Medical Center at Princeton, Princeton, NJ.

4. Results:

a. Table 1 and Figures 1 and I-1 give mean serum levels for aztreonam determined by both the microbiological and HPLC assay procedures.

b. Table 2 and Figures I-4 and 5 give mean cumulative urinary excretion results for aztreonam determined by both the microbiological and HPLC assay procedures. Table 2A gives mean aztreonam urinary excretion concentrations by the bioassay procedure.

c. Table 3 summarizes the pharmacokinetic parameters for aztreonam as determined from both the microbiological and HPLC determined serum levels.

d. Table 4 gives dose proportionality ratios using mean AUC_{0-24} and Cmax values for both microbiological and HPLC serum level results.

e. Figures 2 and I-2 give mean Cmax vs. dose plots and Figures 3 and I-3 give mean AUC_{0-24} vs. dose plots.

5. Comments:

a. Similar results obtained by the microbiological and HPLC assay procedures for mean percent urinary excretion suggest there are little or no bioactive metabolites of aztreonam excreted in urine (Table 2).

b. The major pathway of elimination for aztreonam appears to be via the kidney. Approximately 68 (range 61 to 74%) and 70% (range 57 to 83%) of the administered doses were excreted in urine over 24 hours as determined by the microbiological and HPLC procedures, respectively (Table 2).

c. Serum drug levels of azetreonam are described by an open two compartment kinetic model. The mean (CV) $t_{1/2}$ values (n=36) over all the administered dose levels were 1.66(11) and 1.88(14) for the microbiological and HPLC procedures, respectively.

d. Azetreonam appears to demonstrate dose independent linear pharmacokinetics following single intravenous doses of the drug over the range of [redacted] mg. Mean total body clearance values across all doses were similar (Table 3). AUC_{0-24} dose-proportionality ratios suggest aztreonam to be dose proportional (Table 4) over the studied dosing range.

6. Conclusion:

Study #18,554-1 is an acceptable study in that it describes aztreonam's overall pharmacokinetics, apparent dose proportionality and urinary excretion following single intravenous doses (3 minute infusion) that cover a range of [redacted]. In the sponsor's proposed package insert the lowest and highest doses to be given as single doses within the b.i.d., t.i.d., or q.i.d. dosing regimens for adult patients with normal renal function are 500, 1000 and 2000 mg, respectively.

Mean Serum Concentrations (mcg/ml)

Mean dosage groups

Dose:	500		1000		2000	
Time (Hrs)	Micro	HPLC	Micro	HPLC	Micro	HPLC
0						
0.08	0	0	0	0	0	0
0.17	58.2	57.2	125	152	242	307
0.33	50.7	52.3	103	129	204	258
0.5	40.3	38.4	76.9	98.7	155	199
1.0	32.9	25.9	65.6	82.7	128	165
1.5	23.3	23.9	48.6	52.2	90.7	108
2	17.7	18.2	36.3	41.1	68.6	86.9
3	13.9	14.8	28.6	34.7	54.6	69.2
4	9.4	10.5	19.0	24.3	40.2	47.2
6	6.7	7.7	13.3	16.3	26.4	34.0
8	2.9	3.7	6.0	7.9	12.8	16.5
12	1.3	1.0	2.7	3.6	6.0	7.8
24	0.26	0	0.57	0	1.2	2.0
	0	0	0	0	0	0

Table 2

Mean % Cumulative Urinary Fraction (0-24 Hours)

n = 6 per dosage group

Dose (mg/m²)	Bioassay	HPLC
500	69.2 (8.5)	61.1 (24)
1000	74.1 (6.9)	93.4 (9.1)
2000	67.2 (15)	78.4 (13)

There was no effect on urine pH; values were within the normal range post-drug administration.

TABLE 2A

Concentration of SQ 26,776 in Urine[a]

Bioassay

Time After Infusion, hr	500	1000	2000
	7.0 ± 0.3	13.8 ± 0.7	28 ± 2
0 - 2	1400 ± 200	3000 ± 1200	6500 ± 1100
2 - 4	250 ± 58	710 ± 370	2700 ± 1200
4 - 6	330 ± 57	720 ± 190	1800 ± 520
6 - 8	140 ± 27	300 ± 77	670 ± 230
8 - 12	50 ± 8	70 ± 10	180 ± 38
12 - 16	8.2 ± 1.6	11 ± 3	41 ± 7
16 - 24	1.9 ± 0.4	2.8 ± 0.5	9.8 ± 3.1
% of dose excreted in urine 0 - 24 hr	69 ± 2	74 ± 3	63 ± 3

Table 3

mean Pharmacokinetic Parameters

Dose (mg)	500	1000	2000
C_{max} (mcg/ml)	58.2 (53.8)	125 (153)	242 (307)
AUC_{0-24}	93.3 (96.7) >0.96	191 (231) >0.83	379 (481) >0.79
CL_T ml/min*kg	1.26 (1.16)	1.22 (0.996)	1.26 (0.996)
K_a hr^{-1}	0.24 (0.31)	0.15 (0.22)	0.19 (0.20)
$K_{1/2\beta}$ hr^{-1}	1.76 (2.11)	1.68 (1.85)	1.82 (1.90)
$K_{1,2}$ hr^{-1}	1.01 (1.09)	1.98 (1.35)	1.50 (1.55)
K_{21} hr^{-1}	1.53 (1.55)	2.18 (1.52)	1.71 (1.63)
K_{10} hr^{-1}	0.79 (0.66)	0.88 (0.81)	0.82 (0.82)
V_1 L/kg	0.101 (0.106)	0.083 (0.073)	0.093 (0.074)
V_{ss} L/kg	0.167 (0.179)	0.159 (0.136)	0.175 (0.142)
V_{area} L/kg	0.190 (0.212)	0.177 (0.157)	0.199 (0.163)

* Microbiological Assay

ACTUAL DOSES OF ^{14}C-AZITHROMYCIN ADMINISTERED

Subject No.	IV			IM		
	Log	mg	µCi	Log	mg	µCi
Mean ± S.E.		525.7 ± 10.9	51.9 ± 1.4		511.2 ± 4.5	50.2 ± 0.4

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TABLE 6

CONCENTRATIONS OF TOTAL RADIOACTIVITY, AZITHROMYCIN, SQ 26,992, AND OTHER METABOLITES IN THE SERUM AFTER INTRAVENOUS ADMINISTRATION OF A SINGLE 500-MG DOSE OF ^{14}C-AZITHROMYCIN

TIME (HR)	TOTAL RADIOACTIVITY (µg/ml)[a,c]	CONCENTRATION IN SERUM (µg/ml)[a,c]						
		NONEXTRACTABLE RADIOACTIVITY	EXTRACTABLE RADIOACTIVITY[b]					
			Azithromycin	SQ 26,992	ZONE A	ZONE B	ZONE C	ZONE F
0.083	66.17±3.61	3.66±0.37	43.7±3.03	15.44±2.38	0.37±0.14	0.19±0.07	1.80±0.34	0.97±0.15
0.167	54.15±2.09	2.52±0.31	36.6±1.19	11.50±1.67	0.48±0.19	0.36±0.10	1.62±0.59	0.83±0.15
0.333	44.28±1.92	3.96±0.49	29.3±0.97	8.50±1.21	0.45±0.10	0.28±0.07	1.08±0.23	0.75±0.24
0.5	37.10±1.78	3.31±0.44	24.5±1.03	7.14±0.91	0.26±0.04	0.20±0.03	1.16±0.49	0.50±0.08
1.0	25.03±1.24	2.12±0.24	15.1±1.44	5.13±0.96	0.26±0.05	0.24±0.06	1.65±1.04	0.48±0.07
1.5	19.53±0.82	2.38±0.22	12.1±0.38	3.48±0.63	0.21±0.03	0.16±0.02	0.78±0.29	0.46±0.07
2.0	15.48±0.58	1.90±0.11	9.39±0.31	2.90±0.51	0.18±0.03	0.14±0.03	0.54±0.08	0.35±0.06
3.0	10.90±0.44	1.81±0.13	6.14±0.21	1.92±0.34	0.19±0.04	0.14±0.04	0.32±0.08	0.37±0.06
4.0	7.70±0.30	1.57±0.11	3.90±0.34	1.00±0.26	0.38±0.06	0.13±0.05	0.27±0.06	0.64±0.27
6.0	4.12±0.22	1.40±0.06	1.64±0.11	0.54±0.11	0.07±0.01	0.10±0.03	0.13±0.03	0.25±0.09
8.0	2.53±0.15	1.31±0.07	0.56±0.06	0.26±0.04	0.08±0.02	0.07±0.01	0.05±0.01	0.19±0.09
12.0	1.60±0.06	1.25±0.04	0.08±0.02	0.09±0.02	0.05±0.01	0.02±0.00	0.02±0.00	0.08±0.03
16.0	1.40±0.14	1.21±0.13	0.05±0.01	0.04±0.01	0.02±0.01	0.02±0.00	0.01±0.00	0.05±0.01

a Mean (±S.E.M.) for six subjects.
b See Figure 81 for definitions of Zones A through F
c Equivalents of azithromycin
d Unchanged azithromycin; these values are not corrected for recovery of spiked samples; see Table 12 for adjusted values.
e SQ 26,992 (Major metabolite)

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TABLE 12

CONCENTRATIONS OF AZITHROMYCIN IN SERUM AFTER INTRAVENOUS AND INTRAMUSCULAR ADMINISTRATION OF SINGLE 500-MG DOSES OF ^{14}C-AZITHROMYCIN MEASURED BY BIOASSAY AND RADIOASSAY

Time Hr	Mean (±SEM) Serum Concentrations of Azithromycin, µg/ml			
	Intravenous Dose		Intramuscular Dose	
	Radioassay[a]	Bioassay	Radioassay[a]	Bioassay
0.083	62.0±4.30	54.65±2.77	7.15±2.21	6.04±1.78
0.167	52.0±1.69	45.33±1.72	13.0±3.05	11.19±2.23
0.333	41.5±1.37	37.75±1.68	18.2±2.19	16.54±1.95
0.5	34.8±1.46	32.97±1.68	20.9±3.66	18.93±1.35
1.0	20.0±1.86	19.99±0.74	23.4±1.83	21.27±0.90
1.5	17.1±0.53	15.13±0.69	21.6±1.87	19.73±0.59
2.0	13.3±0.43	12.36±0.46	18.9±1.31	17.38±0.58
3.0	8.72±0.30	8.16±0.41	13.8±0.76	12.76±0.34
4.0	5.56±0.48	6.44±0.27	9.91±0.61	8.72±0.34
6.0	2.33±0.15	2.28±0.16	4.80±0.24	4.09±0.26
8.0	0.80±0.09	0.90±0.08	1.59±0.20	1.70±0.16
12.0	0.11±0.02	0.14±0.02	0.25±0.05	0.30±0.04
16.0	0.07±0.01	0.07±0.01	0.14±0.04	0.04±0.02

a Corrected for recovery of spiked samples (see text for details)

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TABLE 7

CONCENTRATIONS OF TOTAL RADIOACTIVITY, AZITHROMYCIN, SQ 26,992, AND OTHER METABOLITES IN THE SERUM AFTER INTRAMUSCULAR ADMINISTRATION OF A SINGLE 500-MG DOSE OF ^{14}C-AZITHROMYCIN

TIME (HR)	CONCENTRATION IN SERUM (µg/ml)[a,c]							
	TOTAL RADIOACTIVITY (µg/ml)[a,c]	NONEXTRACTABLE RADIOACTIVITY	EXTRACTABLE RADIOACTIVITY[b]					
			Azithromycin	SQ 26,992	ZONE A	ZONE B	ZONE C	ZONE F
0.083	7.75±2.27	0.54±0.22	5.04±1.56	1.47±0.45	0.08±0.04	0.05±0.02	0.22±0.06	0.39±0.17
0.167	13.97±3.17	1.07±0.39	9.17±2.15	2.89±0.68	0.14±0.05	0.06±0.02	0.37±0.13	0.25±0.09
0.333	19.80±2.23	1.41±0.22	12.88±1.54	4.10±0.55	0.13±0.03	0.10±0.02	0.61±0.19	0.58±0.08
0.5	22.90±2.22	1.74±0.14	14.73±2.44	4.87±0.76	0.13±0.02	0.11±0.03	0.77±0.12	0.60±0.11
1.0	26.18±1.08	2.66±0.23	16.49±1.15	5.17±0.84	0.19±0.04	0.14±0.03	0.86±0.25	0.67±0.13
1.5	27.28±1.05	2.01±0.29	15.22±1.32	5.13±1.13	0.28±0.09	0.29±0.05	1.01±0.35	0.64±0.05
2.0	21.32±0.86	2.36±0.20	13.30±0.78	4.04±0.78	0.20±0.06	0.18±0.04	0.72±0.27	0.51±0.10
3.0	16.15±0.54	2.24±0.18	9.74±0.53	2.98±0.56	0.17±0.03	0.13±0.04	0.49±0.14	0.40 0.06
4.0	11.41±0.51	1.80±0.09	6.35±0.29	2.19±0.39	0.18±0.03	0.14±0.04	0.35±0.09	0.41±0.08
6.0	6.07±0.27	1.56±0.05	2.82±0.17	1.17±0.11	0.08±0.01	0.07±0.02	0.11±0.04	0.25±0.06
8.0	3.51±0.18	1.46±0.07	1.12±0.14	0.45±0.07	0.15±0.04	0.06±0.02	0.08±0.02	0.18±0.06
12.0	1.82±0.09	1.28±0.04	0.17±0.03	0.14±0.03	0.06±0.02	0.03±0.01	0.03±0.01	0.12±0.04
16.0	1.46±0.05	1.14±0.05	0.10±0.03	0.07±0.02	0.02±0.01	0.02±0.01	0.02±0.00	0.09±0.02

a Mean (±S.E.M.) for six subjects.
b See Figure 81 for definitions of Zones A through F
c Equivalents of azithromycin
d Unchanged azithromycin; these values are not corrected for recovery of spiked samples; see Table 12 for adjusted values.
e SQ 26,992 (Major metabolite)

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TABLE 9

SUMMARY OF BIOTRANSFORMATION PROFILE OF AZTREONAM IN URINE AND FECES[a]

	Intravenous[b]		Intramuscular[c]	
Number of Subjects	4	6	4	6
Components of Cumulative 0-144 hr Urinary Radioactivity:				
aztreonam	85.9±1.5	87.6±5.6	86.3±2.1	87.1±2.0
SQ 26,992	6.9±0.5	6.9±0.4	7.6±0.4	7.4±0.6
unknowns	3.2±0.2	3.0±0.2	3.8±0.1	3.8±0.1
Components of Cumulative 0-144 hr Fecal Radioactivity:				
aztreonam	1.4±0.0	1.5±0.0	1.9±0.0	0.9±0.1
SQ 26,992	3.4±0.1	3.6±0.1	3.2±0.1	2.9±0.2
unknowns	7.5±0.1	7.8±0.2	10.8±0.4	9.8±0.8
Cumulative Total Urinary and Fecal Excretion 0-144 hr (range)	88.4±1.4 (86.8-92.8)	85.1±5.4 (58.9-92.8)	92.9±2.3 (86.2-96.3)	91.8±1.6 (86.2-96.3)

[a] All values are mean ± S.E.M. as % of total radioactive dose administered. Because of incomplete urine collections for Subjects [redacted] after intravenous dosing, data for Subjects [redacted] were analyzed separately (N=4). For consistency, this was also done for fecal excretion after intravenous dosing, and for all data after intramuscular administration of drug.

[b] Values from Table 846, times 3, 4, 7, 14, 15, 20 and 21.

[c] Values from Table 847, times 3, 4, 7, 14, 15, 7 and 21.

[d] Cumulative urinary excretion 0-48 hrs after IM dosing. No fecal data collected after IM dosing. Fecal data collected after IV dosing only.

Protocol 18554-2

TABLE 9A

INDIVIDUAL VALUES OF PHARMACOKINETIC PARAMETERS FOR AZTREONAM BASED ON SERUM AZTREONAM CONCENTRATIONS MEASURED BY BIOASSAY AND TLRC AFTER INTRAVENOUS ADMINISTRATION OF ^{14}C-AZTREONAM

PARAMETER		SUBJECT NUMBER [redacted]	MEAN ±SEM
$t_{1/2\alpha}$, hr	B		0.25±0.03
	R		0.23±0.03
k_{12}, hr^{-1}	B		1.05±0.20
	R		1.23±0.27
k_{21}, hr^{-1}	B		1.57±0.13
	R		1.60±0.12
V_1, L/kg	B		0.11±0.01
	R		0.09±0.01
V_{ss}, L/kg	B		0.18±0.00
	R		0.16±0.01
V_{area}, L/kg	B		0.21±0.00
	R		0.18±0.01
$t_{1/2\beta}$, hr	B		1.64±0.05
	R		1.56±0.08
k_{10}, hr^{-1}	B		0.81±0.05
	R		0.90±0.08
serum clearance, ml/(min kg)	B		1.50±0.03
	R		1.41±0.03
renal excretion[b], % of dose	B		67.3±2.6
	R		65.9±1.9
renal clearance, ml/(min kg)	B		1.03±0.04
	R		0.94±0.05

[a] B refers to bioassay; R refers to TLRC (radioassay).

[b] Because of incomplete urine collections for Subjects 2 and 3 after intravenous dosing only renal excretion data for Subjects 1, 4, 5, and 6 are analyzed here.

[c] 0 to 48 hours.

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TABLE 9B

INDIVIDUAL VALUES OF PHARMACOKINETIC PARAMETERS FOR AZTREONAM BASED ON SERUM AZTREONAM CONCENTRATIONS MEASURED BY BIOASSAY AND TLRC AFTER INTRAMUSCULAR ADMINISTRATION OF ^{14}C-AZTREONAM

PARAMETER		SUBJECT NUMBER [redacted]	MEAN ±SEM
t_{peak}, hr	B		0.97±0.08
	R		1.00±0.13
$t_{1/2}$, hr	B		0.39±0.13
	R		0.40±0.13
k_0, hr^{-1}	B		2.64±0.59
	R		2.90±0.93
V_{area}, L/kg	B		0.21±0.01
	R		0.19±0.01
$t_{1/2\beta}$, hr	B		1.77±0.08
	R		1.67±0.05
k_{el}, hr^{-1}	B		0.39±0.07
	R		0.41±0.07
serum clearance ml/(min kg)	B		1.40±0.04
	R		1.33±0.06
renal excretion[b], % of dose	B		67.7±4.1
	R		67.0±2.0
renal clearance ml/(min kg)	B		0.95±0.05
	R		0.89±0.04

[a] B refers to bioassay; R refers to TLRC (radioassay).

[b] 0 to 48 hours.

TABLE 9A

BIOAVAILABILITY PARAMETERS FOR AZITHROMYCIN AND TOTAL RADIOACTIVITY IN STRUM OF SUBJECTS AFTER INTRAMUSCULAR OR INTRAVENOUS ADMINISTRATION OF A SINGLE 500-MG DOSE OF ^{14}C-AZITHROMYCIN

ROUTE OF ADMIN.	PARAMETER	SUBJECT NUMBER	Mean ± SEM
I.m.	Cmax (µg/ml)		
	Total Radioactivity		26.8±1.88
	Unchanged Azithromycin		21.5±0.96
I.m.	Tmax (hr)		
	Total Radioactivity		0.92±0.08
	Unchanged Azithromycin		0.92±0.08
I.m.	AUC (µg • hr/ml)		
	Total Radioactivity (0-16 hr)		190.7±3.99
	Unchanged Azithromycin (0-16 hr)		82.4±3.82
I.m.	t1/2(hr)		
	Total Radioactivity (1.5-12 hr)		2.70±0.05
	Unchanged Azithromycin		1.77±0.08
I.v.	Cmax (µg/ml)		
	Total Radioactivity		66.2±3.6
	Unchanged Azithromycin		54.6±2.8
I.v.	Tmax (hr)		
	Total Radioactivity		0.08±0.00
	Unchanged Azithromycin		0.08±0.00
I.v.	AUC (µg • hr/ml)		
	Total Radioactivity (0-16 hr)		116.1±4.96
	Unchanged Azithromycin (0-16 hr)		80.5±3.70
I.v.	t1/2 (hr)		
	Total Radioactivity (1-12 hr)		2.70±0.03
	Unchanged Azithromycin		1.64±0.05
	Absolute Intramuscular Bioavailability of Unchanged Azithromycin. (AUC I.m./AUC I.v.) x 100, %		102.7±2.3 → d.e. / 78.2 ± 2.7 → TLPC

a Total radioactivity is expressed as equivalents of azithromycin; unchanged azithromycin is by bioassay. Results for bioavailability of unchanged azithromycin based on TLPC data are identical and are given in Appendix B, Table B48.

TABLE 7B

BINDING OF ^{14}C-AZITHROMYCIN EQUIVALENTS TO SERUM PROTEIN AT 0.5, 1, AND 3 HOURS FOLLOWING INTRAVENOUS ADMINISTRATION OF A SINGLE DOSE (500 MG) OF ^{14}C-AZITHROMYCIN

SUBJECT NUMBER	TIME (HR)	AZITHROMYCIN EQUIVALENTS IN SERUM (µG/ML)	AZITHROMYCIN EQUIVALENTS IN PFF[a] (µG/ML)	APPARENT BOUND (%)[b]
1	0.5			
1	1.0			
1	3.0			
2	0.5			
2	1.0			
2	3.0			
3	0.5			
3	1.0			
3	3.0			
4	0.5			
4	1.0			
4	3.0			
5	0.5			
5	1.0			
5	3.0			
6	0.5			
6	1.0			
6	3.0			
Mean±SEM	0.5	37.1±2.78	11.1±0.82	69.8±1.79
Mean±SEM	1.0	35.0±1.24	7.45±0.44	78.83±1.95
Mean±SEM	3.0	10.9±0.44	2.88±0.14	73.29±1.74

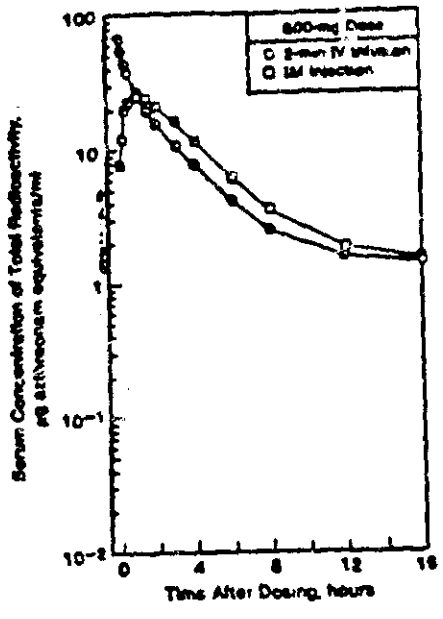
a Protein-free filtrate.
b % Bound = $\frac{\text{Conc. in Serum} - \text{Conc. in PFF}}{\text{Conc. in Serum}} \times 100$

TABLE 7B

BINDING OF ^{14}C-AZITHROMYCIN EQUIVALENTS TO SERUM PROTEIN AT 0.5, 1, AND 4 HOURS FOLLOWING INTRAMUSCULAR ADMINISTRATION OF A SINGLE DOSE (500 MG) OF ^{14}C-AZITHROMYCIN

SUBJECT NUMBER	TIME (HR)	AZITHROMYCIN EQUIVALENTS IN SERUM (µG/ML)	AZITHROMYCIN EQUIVALENTS IN PFF[a] (µG/ML)	APPARENT BOUND (%)[b]
1	0.5			
1	1.0			
1	3.0			
2	0.5			
2	1.0			
2	3.0			
3	0.5			
3	1.0			
3	3.0			
4	0.5			
4	1.0			
4	3.0			
5	0.5			
5	1.0			
5	3.0			
6	0.5			
6	1.0			
6	3.0			
Mean±SEM	0.5	22.9±2.22	5.74±0.49	74.22±1.74
Mean±SEM	1.0	26.2±1.88	7.03±1.00	78.49±2.54
Mean±SEM	3.0	10.2±0.54	2.79±0.17	73.38±0.85

a Protein-free filtrate.
b % Bound = $\frac{\text{Conc. in Serum} - \text{Conc. in PFF}}{\text{Conc. in Serum}} \times 100$



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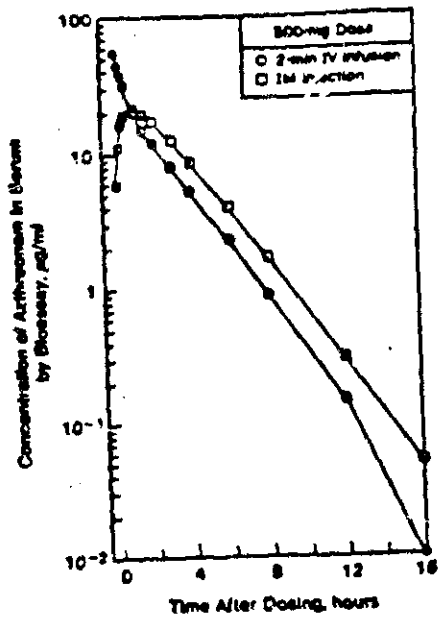

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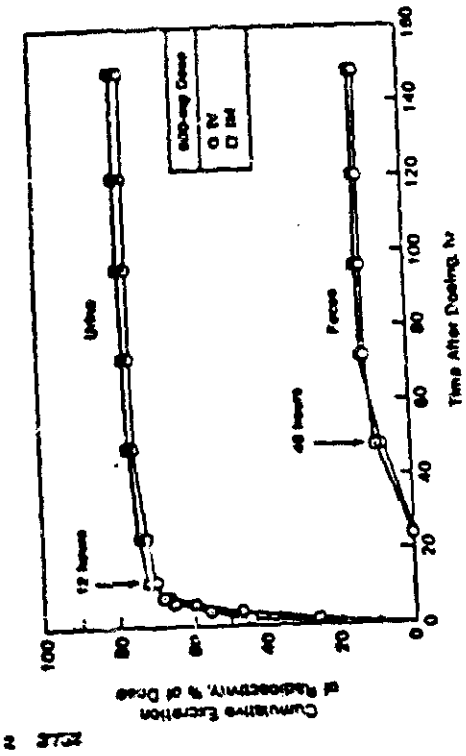

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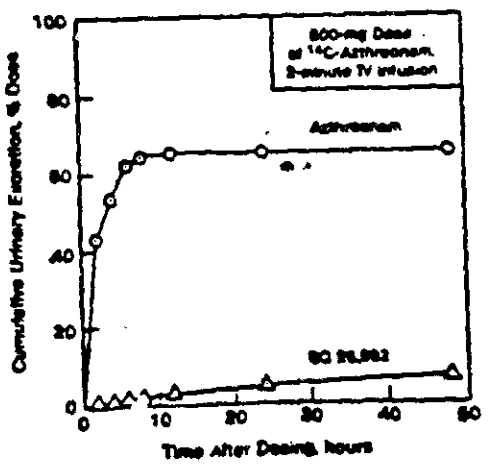

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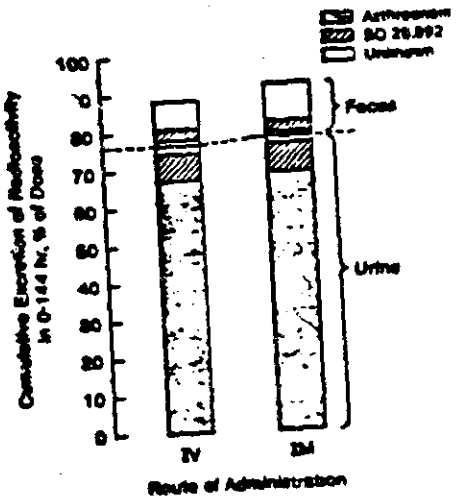

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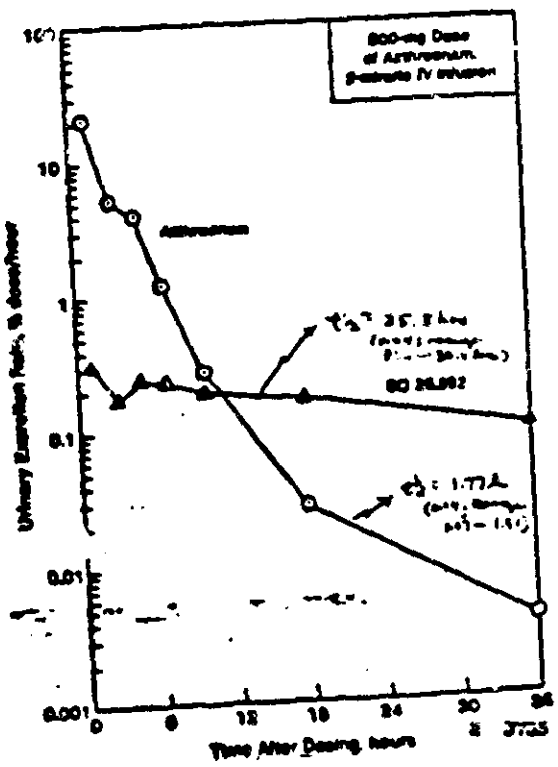

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Poland

C. Study Protocol #18,554-3 (Pivotal Study)

1. Title: Ascending-dose intramuscular safety and pharmacokinetic study of aztreonam in healthy subjects.

2. Objective: Define aztreonam's pharmacokinetics, dose proportionality, urinary excretion and safety following increasing intramuscular doses.

3. Study Design: Twenty-four healthy male subjects (ages 18 to 31 years; mean weight = 73.3 kg) participated in this study. Three groups of 8 subjects each were randomly selected and within each group, 6 subjects were received drug and 2 subjects received placebo. Each group received a different, single, intramuscular dose of aztreonam: 250, 500 or 1000 mg. IM injections were given in the right gluteus maximus muscle. Drug was supplied as a sterile powder blend of aztreonam and L-arginine (ratio=1.0/0.78) which was reconstituted with sterile water. The volume administered per dose was 3.5 ml. 250 ml of water were ingested at the time of drug administration and then at 1 and 2 hours post-dose.

Serial serum and urine samples were collected over 24 and 72 hours, respectively, post-dose. Drug concentrations in serum and urine were determined by the microbiological agar diffusion method. A one-compartment, first-order absorption and elimination model was used to calculate the PK parameters.

This study was conducted by A.A. Sugerman, M.D., the Medical Center at Princeton, Princeton, NJ.

4. Results:

a. Table 1 and Figure 1 gives mean serum drug profiles. Table 1A gives mean urinary excretion concentrations.
b. Table 2 gives summary pharmacokinetic data.
c. Figures 2 and 3 give Cmax vs. dose and AUC_{0-24} vs. dose plots, respectively. Figure 6 gives a cumulative urinary excretion plot.

5. Comments:

a. This study demonstrated single IM doses of aztreonam given over a range of 250, 500 and 1000 mg to be dose proportional. Linear pharmacokinetics were observed in this dosage range.

b. The pharmacokinetic results for the 500 mg IM dose given in this study are consistant with the pharmacokinetic results for the 500 mg IM dose given in Study #18554-2.

6. Conclusion:

Study #18,544-3 is acceptable in that it demonstrated aztreonam to be dose proportional over a range of 250, 500 and 1000 mg when given as single IM injections.

Table 1

Mean (± S.E.) Concentration (mcg/ml)

Dose (mg)

Time (Hr)	500	1000
0.00 HR	0.000	0.000
0.08 HR	4.792	8.613
0.17 HR	8.152	15.753
0.33 HR	15.067	28.233
0.50 HR	18.000	36.050
1.00 HR	21.950	46.217
1.50 HR	19.267	43.167
2.00 HR	16.800	37.867
3.00 HR	12.650	27.100
4.00 HR	8.873	18.400
6.00 HR	3.782	8.233
8.00 HR	1.677	3.538
12.00 HR	0.297	0.646
24.00 HR	0.000	0.000

TABLE ~~XI~~ A

MEAN CONCENTRATIONS (uGM/ML) OF SQ 26,776 IN URINE AFTER SINGLE INTRAMUSCULAR DOSES OF 250, 500, AND 1000 MG

Time After Injection (hrs)	DOSE 500	1000 (mg)
	6.77 ± 0.4	13.98 ± 0.5 (mc/cc
0 - 2	520 ± 190	1200 ± 320
2 - 4	380 ± 170	650 ± 94
4 - 6	420 ± 97	640 ± 200
6 - 8	180 ± 31	470 ± 140
8 - 12	27 ± 8	140 ± 28
12 - 16	6 ± 1	25 ± 4
16 - 24	1.3 ± 0.3	5 ± 2.7
% of dose excreted in urine 0-24h	62 ± 4	69 ± 3

Table 2

Mean (CV) Pharmacokinetic Parameters

n = 6

Dose (mg): Parameter	500	1000
Cmax (mcg/ml)	22 (20)	47.4 (15)
AUC_{0-24} (mcg·hr/ml)	84.1 (9.6)	179.6 (5.5)
Tmax (hr)	1.0 (0)	1.25 (22)
$t_{1/2ab}$ (hr)	0.45 (60)	0.57 (43)
Ka (hr^{-1})	1.98 (48)	1.63 (51)
Vdarea (L/kg)	0.20 (16)	0.19 (10)
$t_{1/2el}$ (hr)	1.67 (19)	1.57 (11)
Kel (hr^{-1})	0.42 (17)	0.45 (11)
% Urinary Exc. (0-24)	61.5 (13)	69.3 (9)

FIGURE 5

MEAN SERUM SO 26,776 CONCENTRATIONS VS. TIME AFTER SINGLE INTRAMUSCULAR DOSES OF 250, 500 AND 1000 mg

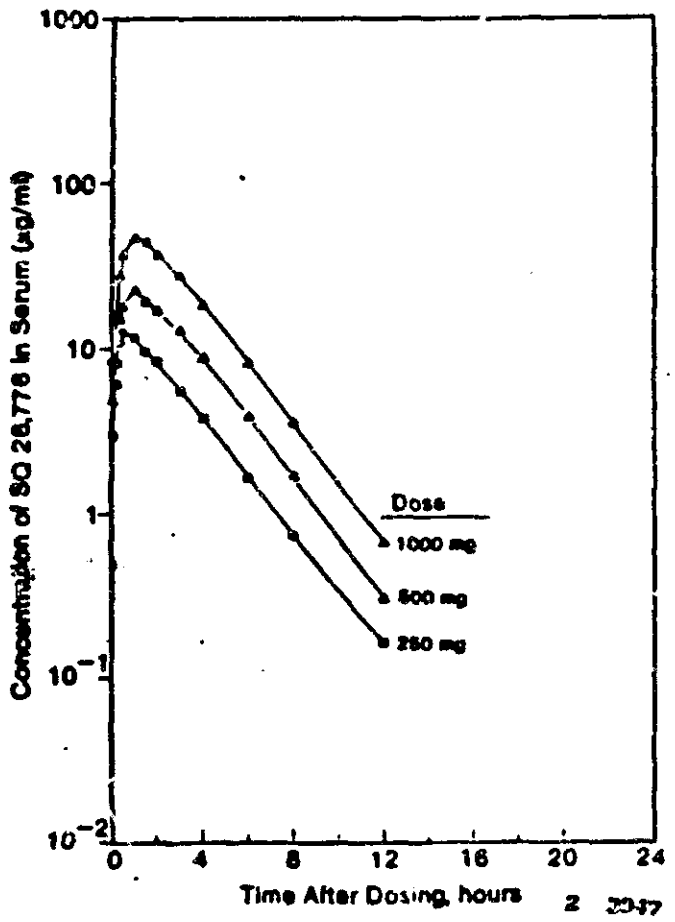

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FIGURE 6

CUMULATIVE URINARY EXCRETION OF SO 26,776 ... TO 72 HOURS AFTER ADMINISTRATION PLOTTED AS PERCENT OF ADMINISTERED DOSE

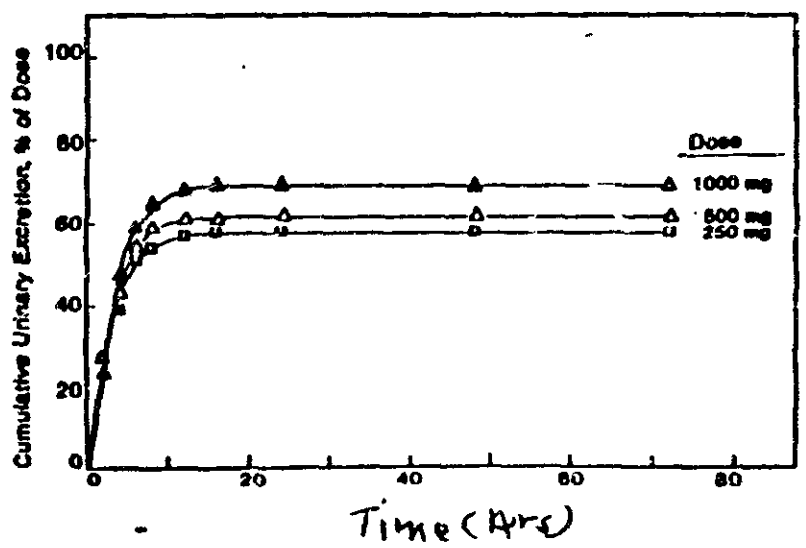


TABLE 2

PEAK SERUM SQ 26,776 CONCENTRATIONS, C_{max} (μg/ml), AFTER THE FIRST AND LAST DOSES OF A SEVEN-DAY QBH INTRAVENOUS DOSING REGIMEN

DOSE					
	500 mg qBh			1000 mg qBh	
Subject No.	C_{max} Day 1	C_{max} Day 8	Subject No.	C_{max} Day 1	C_{max} Day 8
2			14		
3			15		
4			16		
6			18		
7			19		
10			20		
11			21		
12			23		
25			24		
MEAN ± SEM CV	38.7 3.7 29	40.4 4.3 32		99.5 3.0 9	90.1 3.5 12
MEAN (DAYS 1&8) ± SEM	39.6 3.3			95.3 2.6	

TABLE 3

AREA UNDER SERUM SQ 26,776 CONCENTRATION-TIME CURVE (0 TO 8 OR 168 TO 176 HOURS), AUC (μGM x HR/ML), AFTER THE FIRST AND LAST DOSES OF A SEVEN-DAY QBH INTRAVENOUS DOSING REGIMEN

DOSE							
	500 mg qBh				1000 mg qBh		
Subject No.	AUC Day 1	AUC Day 8	Accumulation Ratio	Subject No.	AUC Day 1	AUC Day 8	Accumulation Ratio
2			0.97	14			0.79
3			0.66	15			0.97
4			0.98	16			0.89
6			0.91	18			0.89
7			0.92	19			0.86
10			0.99	20			0.91
11			1.01	21			0.95
12			0.99	23			0.82
25			0.97	24			0.77
MEAN ± SEM CV	68.8 6.8 30	64.0 6.9 34	1.03 — 63		168.4 8.9 11	150.2 8.0 12	0.86 — 9
MEAN (DAYS 1&8) ± SEM	66.4 5.3				159.3 5.8		

TABLE 4

SERUM PROTEIN BINDING (% BOUND) OF SQ 26,776 AFTER THE FIRST AND LAST DOSES OF A SEVEN-DAY QBH INTRAVENOUS DOSING REGIMEN

DOSE					
	500 mg qBh			1000 mg qBh	
Subject No.	% Bound Day 1 [a]	% Bound Day 50	Subject No.	% Bound Day 14	% Bound Day 28
2			14		
3			15		
4			16		
6			18		
7			19		
10			20		
11			21		
12			23		
25			24		
MEAN ± SEM	55.9 0.8	56.9 1.0		55.5 0.8	55.5 0.8
MEAN (DAYS 1&8) [b] ± SEM	56.4 0.6			56.2 0.8	
OVERALL MEAN ± SD	56.3 1.8				

a) Average of values obtained at 10 min, 1 hr, and 3 hr after administration of SQ 26, 776.

b) Average of values obtained at 167.9 hr, 168 hr 10 min, 168 hr, and 171 hr.

Table 4

Mean (CV) Pharmacokinetic Parameters

n = 9 subjects per dosage group

Parameter	Dose: 500 mg tid		1000 mg tid	
	Day 1	Day 8	Day 1	Day 8
$t_{1/2}\alpha$ (hrs)	0.2 (15)	0.18 (33)	0.19 (16)	0.2 (41)
$t_{1/2}\beta$ (hrs)	1.71 (5)	1.54 (6)	1.75 (5)	1.59 (8)
K_{12} (hrs^{-1})	1.41 (34)	1.60 (38)	1.53 (24)	1.39 (43)
K_{21} (hrs^{-1})	1.82 (15)	1.82 (16)	1.71 (14)	1.76 (27)
V_1 (L/kg)	0.13 (46)	0.12 (50)	0.10 (0)	0.11 (27)
V_{ss} (L/kg)	0.23 (52)	0.23 (52)	0.19 (16)	0.19 (16)
$V_{d_{area}}$ (L/kg)	0.26 (46)	0.25 (48)	0.21 (14)	0.22 (14)
K_{10} (hr^{-1})	0.80 (11)	0.97 (15)	0.86 (7)	0.88 (17)
% Urinary Excretion 0-8 hrs	51.3 (29)	57.8 (22)	66.2 (7)	63.9 (19)
% Urinary Excretion 8-24 hrs	—	59.1 (22)	—	65.9 (18)
C_{lT} (ml/min·kg)	1.74 (50)	1.91 (55)	1.42 (1)	1.61 (13)

FIGURE 1

MEAN SERUM SQ 26,776 CONCENTRATIONS vs TIME DURING DOSAGE REGIMENS OF 500 and 1000 mg IV q8h (n = 9, each regimen)

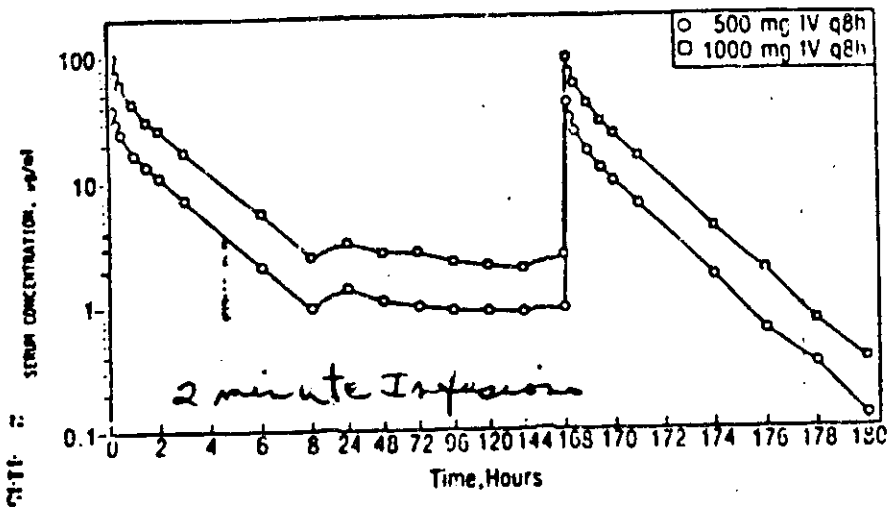
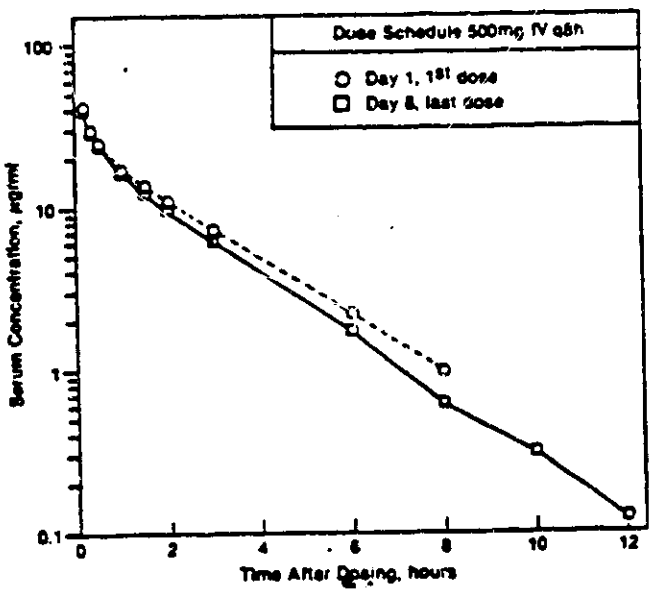


FIGURE 2

COMPARISON OF MEAN SERUM SQ 26,776 CONCENTRATIONS vs TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 500-mg IV DOSES



COMPARISON OF MEAN SERUM SQ 26,776 CONCENTRATIONS vs TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 1000-mg IV DOSES

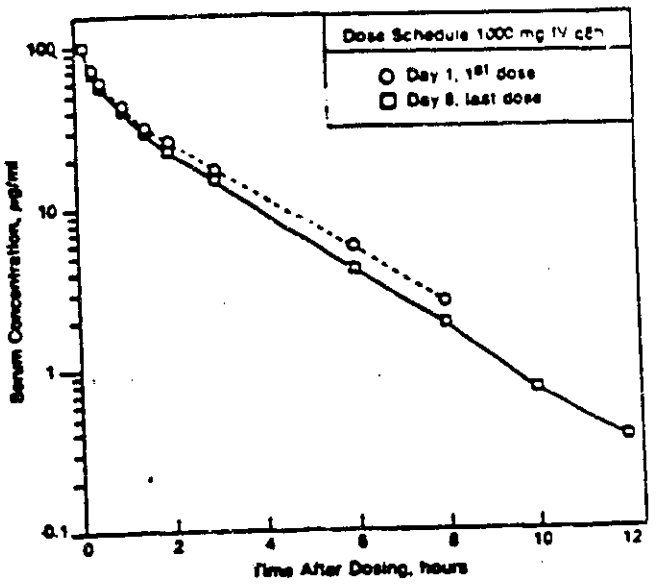


FIGURE 7

COMPARISON OF MEAN CUMULATIVE URINARY EXCRETION OF SQ 26,776 vs TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 500-mg IV DOSES

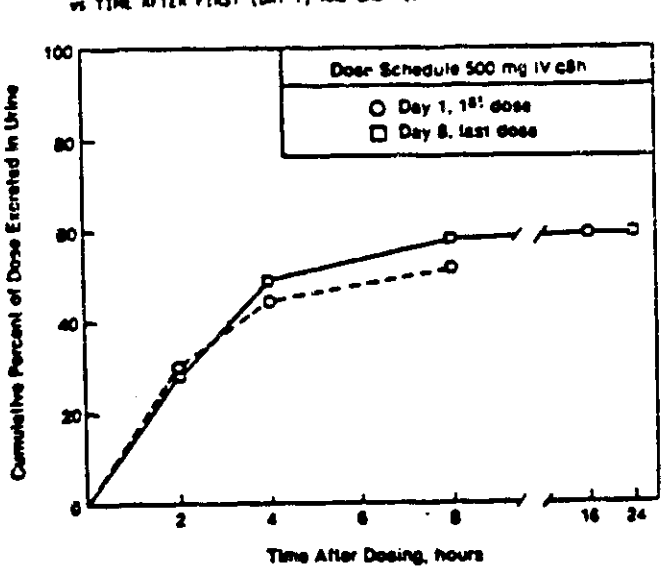
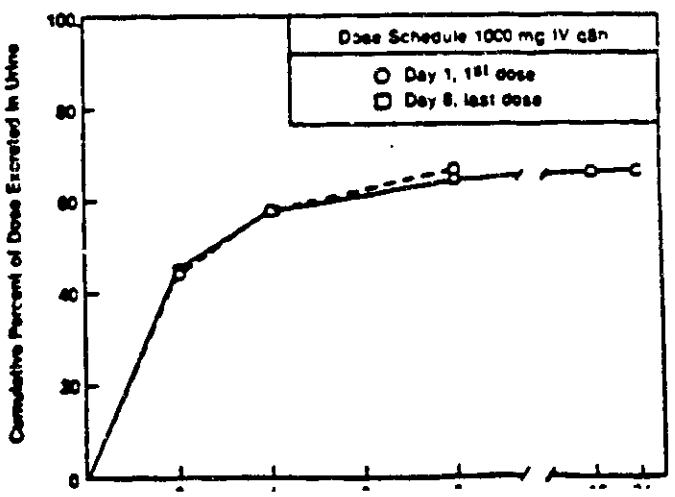


FIGURE 8

COMPARISON OF MEAN CUMULATIVE URINARY EXCRETION OF SQ 26,776 vs TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 1000-mg IV DOSES



E. Study Protocol #18,554-5 (Pivotal Study)

1. Title: Multiple-dose intramuscular safety and pharmacokinetic study of azthreonama in healthy subjects.

2. Objective: The purpose of this study was to determine the safety and pharmacokinetics of aztreonam given IM under multiple dose conditions.

3. Study Design: Enrolled in this study were 24 healthy male subjects (ages between 18 to 30 years; mean weight = 76.2 kg) from which two groups of 12 subjects each were randomly selected. Within each group 9 subjects were given drug and 3 subjects were given placebo. The first study group received aztreonam intramuscularly as 500 mg t.i.d. for 22 doses and the second study group received aztreonam intramuscularly as 1000 mg t.i.d. also for 22 doses. Each dose was injected into the gluteus maximus muscle with each dose being alternated between the right and left sides.

Approximately 250 ml of water were ingested at 0, 1, 2, 168, 169 and 170 hours and water was allowed ad lib during the remainder of the study. Subjects abstrained from eating for at least 8 hours before and 4 hours after the first and last doses at 0 and 168 hours.

Drug was supplied as a sterile powder blend of aztreonam and L-arginine (ratio = 1.0/0.7) which was reconstituted with sterile water to a final volume of 3.5 ml.

Serial serum and urine samples were collected over 180 and 192 hours, respectively. Drug concentrations in serum were determined by the microbiological assay method and drug concentrations in urine were determined both by the microbiological method and a HPLC method. For the high dosing regimen urine major metabolite (SQ 26,992; the open beta lactam ring hydrolysis product of aztreonam) levels were determined using the HPLC method.

This study was conducted by A.A. Sugerman, M.D., the Medical Center at Princeton, Princeton, NJ.

4. Results:

a. Table 1 gives average (SE) serum aztreonam concentrations for the 500 mg and 1000 mg t.i.d. IM dosing schedules. Table 1A gives mean aztreonam urinary excretion concentrations for each dosing regimen.

b. Table 2 gives individuals' Cmax values for Day 1 and Day 8 for each dose level. Table 3 gives similar results for AUC values.

c. Table 4 summarizes the mean generated pharmacokinetic parameters for each dose level for Study Days 1 and 8. Table 5 gives cummulative amounts of aztreonam and its major metabolite (SQ 26,992) excreted in urine for the 1000 mg t.i.d. IM dosing schedule.

d. Figure 1 gives mean serum aztreonam concentration vs. time profiles for both t.i.d. dosing schedules. Figures 2 and 3 give Day 1 vs. Day 8 mean serum concentration vs. time profiles for each dosage level. Figure 7 compares aztreonam urinary levels by both the bioassay and HPLC methods. Figure 9 plots cummulative urinary excretion data for aztreonam's major metabolite as does Figure 10.

5. Comments:

a. No aztreonam accumulation occurs following the IM dosing schedules of 500 mg t.i.d. and 1000 mg t.i.d. given for seven days (22 doses). However, there does appear to be some accumulation of aztreonam's major metabolite (SQ 26,992) based upon urinary excretion data. This would be expected based upon the results from Study Protocol #18, 544-2 which indicated the half-life of the major metabolite to be about 25 hours.

The firm states the following:

The extent of elimination of SQ 26,992 is shown for the high-dose regimen in Figure 9 as cumulative percent of administered azthreonam dose. There was a marked difference between the amount of SQ 26,992 excreted in the urine after the first and last doses. Within 8 hours after the first dose, an average of 1.5% of the dose was excreted in the urine as SQ 26,992, whereas within the same time period after the last dose, an average of 8.4% of the dose was excreted in the urine as SQ 26,992. This suggests that 1) SQ 26,992, unlike azthreonam, was accumulating in the body during the 7-day dosage regimen and/or, 2) the biotransformation of azthreonam to SQ 26,992 is to some extent inducible with pro'onged exposure. In addition, by 24 hours after the last dose, an average of 15.8% of the 1000-mg dose was found in the urine as SQ 26,992 with the curve not yet exhibiting a platesu at that time. Again, this suggests accumulation of SQ 26,992 occurred during the study. In comparison, during the 24-hour period prior to the last dose (144 to 168 hours, Day 7), an average of 5.4% of the total dose (3000 mg over a 24-hour period) was found in the urine as SQ 26,992. The difference between 15.8% and 5.4% appears to represent elimination of SQ 26,992 from an unknown body reservoir during the unsteady-state condition existing after the termination of q8h azthreonam dosing. The 24-hour excretion of SQ 26,992 immediately prior to the last dose represents steady-state conditions, where the average body content of SQ 26,992 is relatively constant.

2 1627

The cumulative excretion of azthreonam and SQ 26,992 is summarized for key collection periods in Table 43. Differences between values of azthreonam measured by bioassay in various collection periods were within experimental variation, and similarly for azthreonam values measured by HPLC assay. In contrast, the four-fold difference in 8-hour SQ 26,992 values on Days 1 and 8, and the three-fold difference in 24-hour SQ 26,992 values on Days 7 and 8 are clearly evident, suggesting accumulation of SQ 26,992 during the seven-day q8h azthreonam dosing regimen.

Figure 10 shows the mean SQ 26,992 in various urine collections expressed as percent of total drug (azthreonam and SQ 26,992, HPLC assay) after the first and last doses of the high-dose regimen. It is clear that the proportion of total drug represented by SQ 26,992 increased with time after dose, and reached 100% in the 16 to 24 hour collection period. This suggested that urinary elimination of SQ 26,992 was slower than azthreonam, either intrinsically, or due to a drug interaction, such as competition for a secretion pump for organic acids. [9]

Additionally the sponsor indicated the following:

While azthreonam did not appear to accumulate during the q8h seven-day dosage regimens, evidence was found for the production and possible accumulation of SQ 26,992, the open, beta-lactam ring hydrolysis product of azthreonam. SQ 26,992 appeared to be excreted in the urine more slowly than azthreonam, thereby possibly leading to accumulation of SQ 26,992. There were two possible sources of SQ 26,992: 1) SQ 26,992 present as an impurity in the azthreonam clinical supplies, and 2) biotransformation of azthreonam by hydrolysis of the beta-lactam ring in vivo to produce SQ 26,992. The SQ 26,992 content

of the azthreonam administered was 0.15 to 0.3% by weight[8], which is less than the mean value of 1.5 ± 0.2% for SQ 26,992 recovery in the urine after the first dose of azthreonam. Biotransformation may explain the appearance of some SQ 26,992 detected in the urine on Day 1. However, on Day 7, under steady-state conditions, 5.4% of the 24-hour dose was found as SQ 26,992 in the urine. This clearly exceeded the amount of SQ 26,992 impurity in azthreonam clinical supplies. Thus, azthreonam undergoes biotransformation to SQ 26,992 in healthy male subjects. The prolonged excretion of SQ 26,992 after the last azthreonam dose suggests that a significant body reservoir exists where SQ 26,992 can accumulate. One may speculate that tissue binding or enterohepatic recirculation could represent such a reservoir. In addition, increased recovery during 8-hour post-dose periods late in the dosage regimen compared to after the first dose is consistent with increased serum levels of SQ 26,992, and/or inducible biotransformation of azthreonam to SQ 26,992 with prolonged exposure to azthreonam. Future multiple-dose studies in patients receiving azthreonam may provide opportunities to confirm these findings. [9]

(*Described in Squibb Analytical R&D Report, dated February 9, 1982. One-gram vials of azthreonam from Lots No. MNB-864-H/B02 and MNB-864-H/B08 were assayed. These supplies came from the same bulk lot of azthreonam as that administered to subjects in this study. Differing terminal digits indicate different filling days for the vials).

b. The firm stated the following regarding renal function assessments.

"No significant alterations in renal function were observed during this study. The standard tests of renal function (serum creatinine, BUN, urinary creatinine clearance), as well as more sensitive enzyme and protein excretion tests (NAG, AAP, and β_2M), gave no indication of renal injury at doses as high as 1000 mg q8h for 7 days.

6. Conclusion:

Study #18,544-5 is an acceptable study in that it defines aztreonam's pharmacokinetics following multiple IM doses of the drug given according to two of the dosage regimens that are to be recommended in the products' package insert (0.5 g and 1.0 g t.i.d.).

Table 1

Average (S.E.) azithromycin serum concentration (mcg/ml).

(n = 9 subjects per dosage level)

500 mg tid

	AVERAGE	S.E.
0.00 HR	0.000	0.000
0.17 HR	8.597	1.174
0.33 HR	13.172	0.962
0.50 HR	15.227	1.023
1.00 HR	18.256	0.982
1.50 HR	17.267	0.667
2.00 HR	15.167	0.551
3.00 HR	11.609	0.447
6.00 HR	4.304	0.278
8.00 HR	2.009	0.178
23.90 HR	2.096	0.279
47.90 HR	1.722	0.[illegible]1
71.90 HR	1.562	0.[illegible]5
95.90 HR	1.807	0.180
119.90 HR	1.589	0.207
143.90 HR	1.798	0.247
167.90 HR	1.714	0.231
168.17 HR	11.489	1.396
168.33 HR	16.644	1.689
168.50 HR	18.956	1.601
169.00 HR	20.178	0.924
169.50 HR	17.556	0.621
170.00 HR	15.500	0.629
172.00 HR	10.836	0.533
174.00 HR	3.519	0.271
176.00 HR	1.457	0.099
178.00 HR	0.607	0.051
180.00 HR	0.247	0.013

1000 mg tid

AVERAGE	S.E.
0.000	0.000
11.722	1.816
21.244	2.713
28.044	3.087
36.111	3.000
34.689	2.150
34.844	1.725
26.611	1.495
9.742	1.008
4.506	0.611
5.867	0.707
4.160	0.3[illegible]
3.931	0.29[illegible]
3.280	0.351
2.901	0.223
2.966	0.268
2.786	0.299
18.983	2.576
31.289	3.590
36.733	3.253
40.344	2.254
36.411	1.[illegible]42
30.844	1.497
21.767	0.980
6.137	0.292
2.497	0.180
0.948	0.103
0.356	0.041

TABLE ~~3~~ 1A

CONCENTRATION OF AZTHREOKAM IN URIN[a] AFTER THE FIRST AND LAST DOSES OF A SEVEN-DAY QBH INTRAMUSCULAR DOSING REGIMEN

	DOSE			
	500 mg QBH		1000 mg QBH	
Time after infusion, hr.	Urine Concentration		Urine Concentration	
	Day 1	Day 8	Day 1	Day 8
0- 2	528 ± 102	969 ± 182	805 ± 135	1996 ± 644
2- 4	370 ± 91	469 ± 96	629 ± 134	835 ± 365
4- 8	283 ± 47	342 ± 44	720 ± 139	428 ± 54
8-16	---	32 ± 6	---	56 ± 11
16-24	---	2 ± 0	---	3 ± 0

[a]Mean values, ± S.E.M., in ug/ml for cumulative urine

TABLE 3?

MAXIMUM SERUM AZTHREONAM CONCENTRATIONS, C_{max} (µg/ml), AFTER THE FIRST AND LAST DOSES OF A SEVEN-DAY QOH INTRAMUSCULAR DOSING REGIMEN

DOSE					
	500 mg q8h			1000 mg q8h	
	C_{max}			C_{max}	
Subject No.	Day 1	Day 8	Subject No.	Day 1	Day 8
2			14		
3			15		
4			16		
5			18		
6			19		
7			20		
10			21		
11			23		
12			24		
MEAN ± SEM CV	18.4 ± 0.9 14.7	21.1 ± 1.2 17.1		38.7 ± 1.8 14.7	41.1 ± 2.2 16.8
MEAN (DAYS 1&8) ± SEM	19.8 ± 0.8			39.9 ± 1.8	

Table 4
Mean (CV) Pharmacokinetic Data
n = 9 subjects per dosage group

Parameter	500 mg q8h Dose		1000 mg q8h Dose	
	Day 1	Day 8	Day 1	Day 8
T_{max} (hr)	1.11(19)	0.92(39)	1.22(42)	1.0 (42)
$t_{1/2\alpha}$ (hr)	0.39(35)	0.27 (44)	0.41(50)	0.24(58)
k_a (hr^{-1})	2.40(53)	3.11(57)	1.86(44)	2.36(53)
$t_{1/2}$ el (hr)	2.08(9)	1.74(5)	1.72(7)	1.57(10)
Kel (hr^{-1})	0.33(4)	0.40(8)	0.40(8)	0.44(14)
V_{area} (L/kg)	0.26(12)	0.22(14)	0.21(14)	0.22(14)
Clt (ml/min/kg)	1.46(7)	1.44(8)	1.43(11)	1.63(17)

TABLE 38

AREA UNDER SERUM AZTHREONAM CONCENTRATION-TIME CURVE (0 TO 8 OR 168 TO 176 HOURS), AUC (µGM x HR/ML), AFTER THE FIRST AND LAST DOSES OF A SEVEN-DAY QOH INTRAMUSCULAR DOSING REGIMEN

DOSE						
	500 mg q8h			1000 mg q8h		
	AUC			AUC		
Subject No.	Day 1	Day 8	Subject No.	Day 1	Day 8	Ratio
			hr)			
2			0.90 14			0.87
3			0.81 15			0.87
4			0.90 16			0.91
5			1.09 18			0.82
6			0.85 19			0.96
7			0.88 20			0.76
10			0.92 21			0.86
11			0.99 23			0.89
12			0.95 24			0.79
MEAN ± SEM CV	74.4 ± 2.3 9.3	73.6 ± 2.7 11.0	0.92 9.9	159.4 ± 6.9 12.0	143.7 ± 5.9 12.7	0.90 7.4
MEAN (DAYS 1&8) ± SEM	74.0 ± 2.4			151.6 ± 6.1		

*Uses 167.9-hour serum concentration as estimate of 168-hour (moment of drug administration) value for AUC calculation.

E3

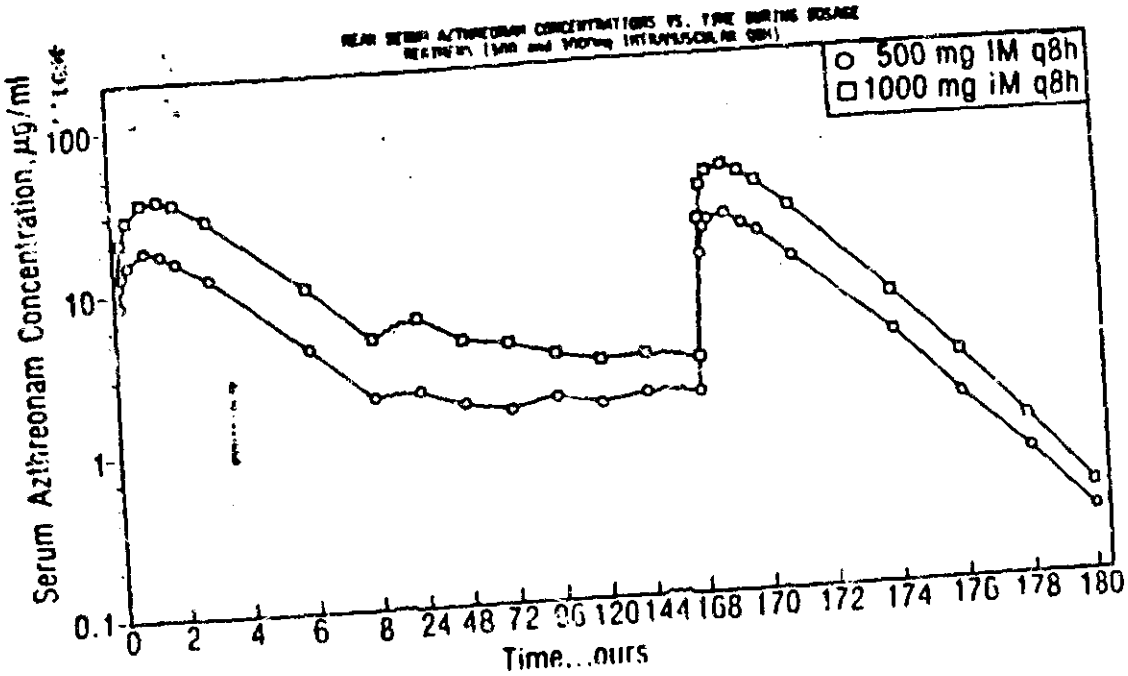


FIGURE 2

COMPARISON OF MEAN SERUM AZTHREONAM CONCENTRATIONS VS. TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 500-mg INTRAMUSCULAR DOSES

FIGURE 3

COMPARISON OF MEAN SERUM AZTHREONAM CONCENTRATIONS VS. TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 1000-mg INTRAMUSCULAR DOSES

TABLE 9

CUMULATIVE AMOUNTS[a] OF AZTHREONAM AND SQ 26,992 EXCRETED IN URINE DURING 1000-mg IM DOSAGE REGIMEN

Collection Period (Duration)	Dose and Dosing Time	Percent of Dose Recovered in Urine As		
		Aztreonam		SQ 26,992
		Bioassay	HPLC	HPLC
0- 8 hr, Day 1 (8 hr)	1 gm at 0 hr	62.6 ± 2.9	52.8 ± 2.4	1.5 ± 0.2
144-168 hr, Day 7 (24 hr)	1 gm at 144, 152 & 160 hr	57.0 ± 5.0	51.2 ± 3.7	5.4 ± 0.5
168-176 hr, Day 8 (8 hr)	1 gm at 168 hr	71.3 ± 5.4	63.2 ± 4.2	6.4 ± 0.8
168-192 hr, Day 8 (24 hr)	1 gm at 168 hr	74.4 ± 5.3	66.3 ± 4.1	15.8 ± 1.3

[a]Mean values ± S.E.M. as percent of dose indicated for cumulative urine collection over indicated time interval.

BIOASSAY AND HPLC ASSAY OF AZTREONAM IN INDIVIDUAL URINE COLLECTIONS DURING THE 1000-mg INTRAMUSCULAR DOSE AZTREONAM DOSING REGIMEN

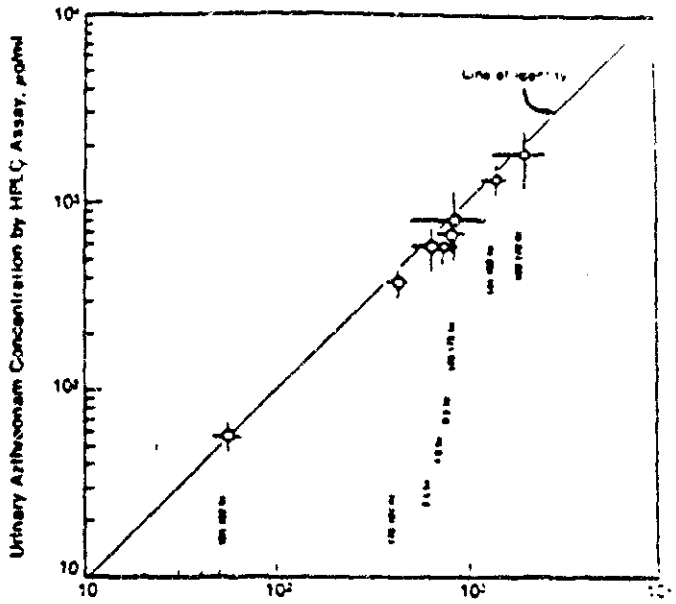


FIGURE 9

COMPARISON OF MEAN (± SEM) CUMULATIVE URINARY EXCRETION OF SQ 26,992 (HPLC ASSAY) VS. TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 1000-mg INTRAMUSCULAR DOSES

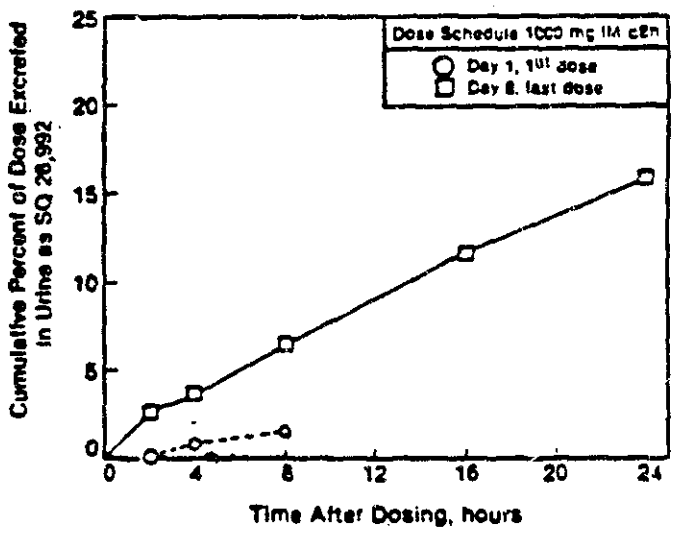
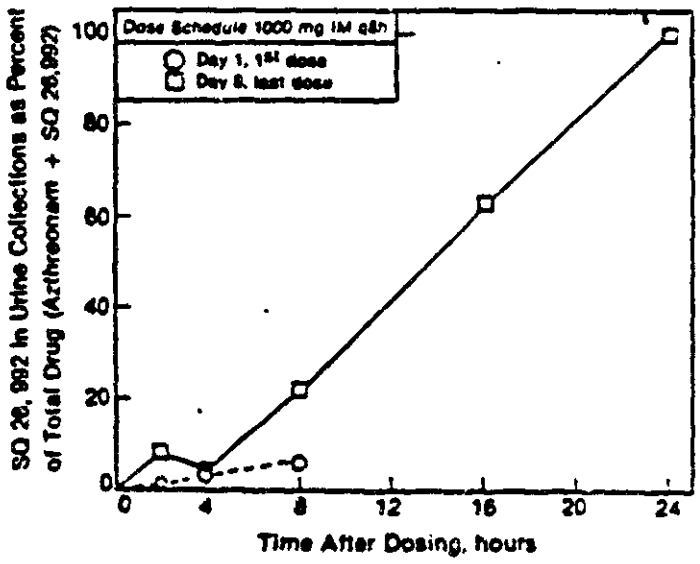


FIGURE 10

COMPARISON OF MEAN (± SEM) SQ 26,992 IN URINE COLLECTIONS AS PERCENT OF TOTAL DRUG (AZTHREONAM PLUS SQ 26,992 - HPLC ASSAY) VS. TIME AFTER FIRST (DAY 1) AND LAST (DAY 8) 1000-mg INTRAMUSCULAR DOSES



F. Study Protocol #18, 544-18) (Supportive Study)

1. Title: Intravenous safety and pharmacokinetic study of aztreonam in healthy subjects (30 min infusion).

2. Objective: The purpose of this study was to evaluate the safety and pharmacokinetics of aztreonam after IV infusion.

3. Study Design: Enrolled in this study were 6 healthy male volunteers (ages between 22-32 years; mean weights=70.1 kg). Each subject received single 500, 1000 and 2000 mg doses of aztreonam given as 30 minute IV infusions in a randomized crossover study design. There was a 1 week wash-out period between each dose. In order to promote urine formation 250 ml of tap water were given to each subject 1 and 2 hours after drug administration.

Drug was supplied as a sterile powder blend of aztreonam and L-arginine (ratio 1.0/0.7) which was reconstituted in sterile water and diluted with 5% sterile dextrose to a final volume of 30 ml. This volume was infused at a rate of 1 ml/min for 30 minutes using a calibrated syringe pump. Serum and urine samples were serially collected over 12.5 hours after the start of drug infusion. Drug concentrations in serum and urine were determined by microbiological assays by both the principal investigator and Squibb Institute. (Note: Only results from Squibb were provided. Squibb indicated the determined PK results by both laboratories were similar for all practical purposes).

The investigator for this study was Harold C. New, M.D. at the College of Physicians and Surgeons of Columbia University, NY, NY.

4. Results:

a. Tables 1 and 2 give mean serum drug concentrations as determined by Squibb and the study's investigator, respectively. Table 3 gives summary pharmacokinetic parameters for each dose.

b. Tables 12 and 13 give individuals' Cmax and AUC_{0-12} values.

c. Figure 1 gives semi-log plots of mean serum level vs. time data data for each dose. Figures 2 and 3 give Cmax vs. Dose and AUC_{0-12} vs. Dose plots, respectively. Figure 4 plots mean cummulative aztreonam urinary excretion data.

5. Comments:

a. Overall aztreonam serum levels determined by Squibb's bioassay were lower, especially at the higher doses of 1000 and 2000 mg, than the drug serum levels determined by the clinical investigator's bioassay. This difference was not observed for determined urine drug concentrations.

b. Comparisons of AUC_{0-12} values and pharmacokinetic parameters for each 30 minute intravenous infusion dose indicate that linear pharmacokinetics appear to prevail over this study's dosage range. The determined pharmacokinetic parameters from this study and Study #18,554-1, where the same intravenous doses were given in 3 minutes, are similar.

6. Conclusions:

Study #18,544-18 is an acceptable study in that it defines aztreonam's pharmacokinetics following single 30 minute intravenous infusions for the three dose levels that are to be given as single doses within the b.i.d. t.i.d. or q.i.d. dosing regimens that are recommended in the product's proposed package insert (i.e., 0.5, 1.0 and 2.0 grams).

SERUM CONCENTRATIONS (MEAN ± S.E.M., MCM/ML) OF AZTHREONAM AFTER SINGLE 30-MIN INTRAVENOUS INFUSIONS[a]

TIME AFTER START OF INFUSION (HR)	DOSE, MG 500	1000	2000
0.5 0	54.0 ± 8.7	90.3 ± 9.9	204 ± 19
0.75 0.25	36.3 ± 5.4	64.8 ± 6.2	135.5 ± 9.0
1.0 0.5	29.7 ± 4.8	52.3 ± 4.9	112.4 ± 8.1
1.5 1	18.9 ± 1.3	38.5 ± 4.5	75.3 ± 8.6
2.0 1.5	15.3 ± 1.1	37.6 ± 4.7	62.7 ± 8.3
2.5 2	12.8 ± 0.9	28.2 ± 2.2	55.3 ± 4.4
4.5 4	5.92 ± 0.67	13.2 ± 1.3	25.5 ± 2.6
6.5 6	2.68 ± 0.40	5.85 ± 0.82	11.7 ± 1.5
8.5 8	1.34 ± 0.14	2.87 ± 0.37	5.79 ± 0.84
12.5 12	0.25 ± 0.05	1.15 ± 0.54	1.39 ± 0.24

[a]Bioassay by The Squibb Institute.

Protocol 18554-18

TABLE 12

MAXIMUM SERUM CONCENTRATIONS[a], C_{max} (MCM/ML), OF AZTHREONAM AFTER SINGLE 30-MIN INTRAVENOUS INFUSIONS

SUBJECT NO.	DOSE, MG 500	1000	2000
1	2.3	2.4	5.5
2	1.9	3.4	2.4
3	[redacted]	2.0	2.1
4	1.9	1.9	3.6
5	1.9	2.1	3.9
6	2.9	2.2	6.3
MEAN ± S.E.M.	54.0 ± 8.7	90.3 ± 9.9	3.8

[a]Bioassay by The Squibb Institute.

Table 2 (Investigator)

	500 AVERAGE	S.E.	1000 AVERAGE	S.E.	2000 AVERAGE	S.E.
0.00 HR	65.460	6.831	163.668	3.134	254.897	22.778
0.25 HR	42.480	4.057	116.752	7.838	200.332	10.748
0.50 HR	32.537	2.177	72.906	4.133	155.285	13.787
1.00 HR	23.108	1.266	48.788	5.699	111.112	13.786
1.30 HR	17.662	1.467	47.283	4.806	76.837	8.834
2.00 HR	13.800	0.626	35.067	2.778	66.855	8.882
4.00 HR	6.870	0.278	16.158	1.850	35.543	2.514
6.00 HR	3.493	0.349	8.478	1.217	14.637	1.431
8.00 HR	1.753	0.327	3.023	0.564	8.545	0.878
12.00 HR	0.172	0.110	0.823	0.137	1.870	0.235

TABLE 13

AREA (TRAPEZOIDAL RULE) UNDER SERUM CONCENTRATION[a]-TIME CURVE, AUC 0-12 hr (uGM X HR/ML), OF AZTHREONAM AFTER SINGLE 30-MIN INTRAVENOUS INFUSIONS

SUBJECT NO.	DOSE, MG 500	1000	2000
1	3.2	3.0	4.6
2	1.6	2.1	3.3
3	1.6	1.9	2.9
4	2.1	1.8	3.2
5	2.0	2.0	4.1
6	3.2	2.2	7.0
MEAN ± S.E.M.	94.7 ± 10.6	182.1 ±15.4	385.1 ± 32.3
GEOM. MEAN	92.2	189.0	378.8

[a]Bioassay by The Squibb Institute.

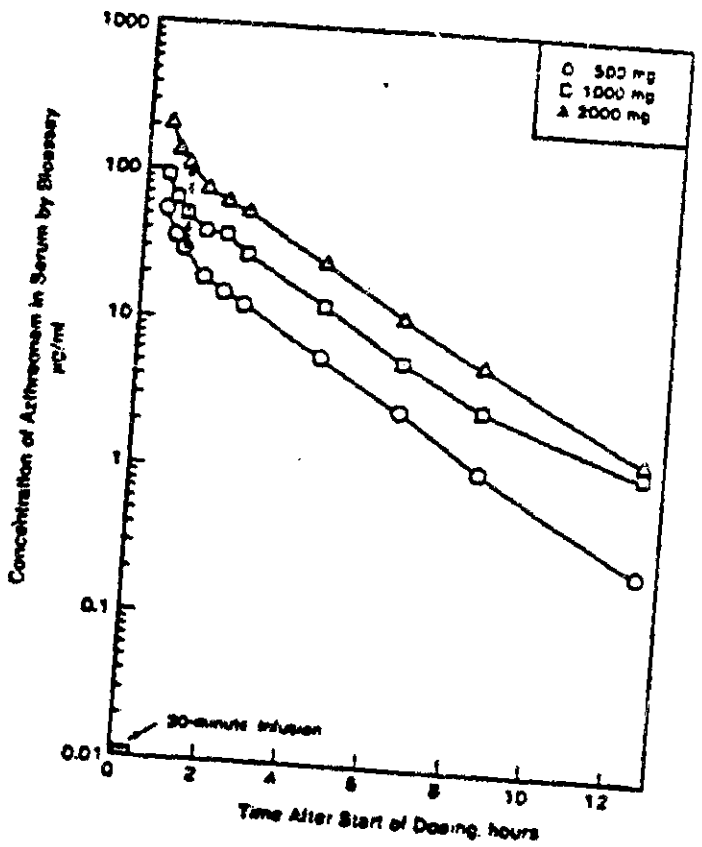
Table 3

Summary Pharmacokinetic Parameters (Mean; C.V.) 30 min. Intravenous Infusion (n=6 per dose)

Parameter	Dose 500 mg	1000 mg	2000 mg
$t_{1/2\alpha}$ (hr^{-1})	0.2 (24)	0.22 (20)	0.20 (24)
$t_{1/2\beta}$ (hr)	1.79 (10)	1.92 (13)	1.90 (12)
K_{12} (hr^{-1})	1.40 (23)	1.25 (36)	1.56 (44)
K_{21} (hr^{-1})	1.62 (29)	1.57 (21)	1.60 (29)
V_1 (L/Kg)	0.09 (27)	0.10 (22)	0.09 (27)
V_{ss} (L/Kg)	0.18 (27)	0.18 (0)	0.18 (14)
V_{area} (L/Kg)	0.20 (24)	0.22 (20)	0.21 (23)
K_{10} (hr^{-1})	0.88 (12)	0.77 (32)	0.89 (38)
Cl_T (ml/min·kg)	1.33 (22)	1.31 (21)	1.29 (19)
Cl_R (ml/min·kg)	0.76 (13)	0.87 (14)	0.81 (15)
% Urinary Exc.	59.5 (23)	67.3 (9)	63.4 (6)

FIGURE

MEAN SERUM AZTHREONAM CONCENTRATIONS VS. TIME AFTER THE START OF 30-MIN INTRAVENOUS INFUSIONS OF 500, 1000, AND 2000 MG OF AZTHREONAM (Assay by Squibb Institute)



MEAN AREA UNDER SERUM AZTHREONAM CONCENTRATION (BIOASSAY) - TIME (0-12 HR) CURVE VS. DOSE OF AZTHREONAM ADMINISTERED AS A 30-MIN INTRAVENOUS INFUSION (Assay by Squibb Institute)

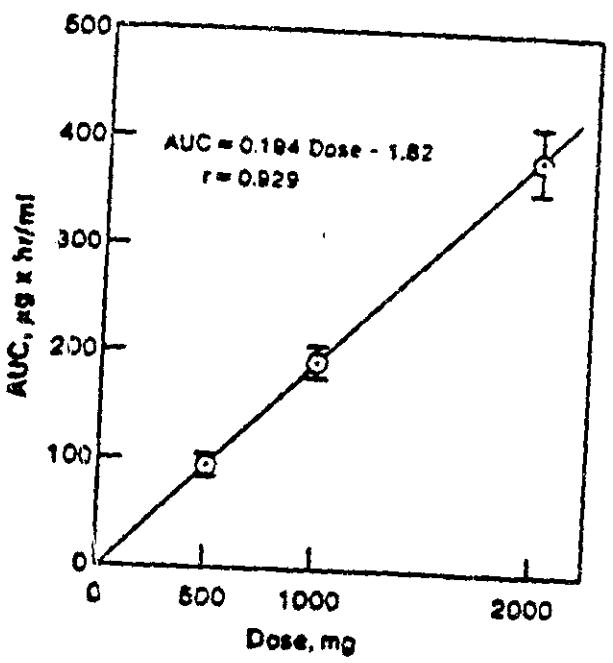


FIGURE 2

MEAN MAXIMUM SERUM AZTHREONAM CONCENTRATIONS BY BIOASSAY VS. DOSE OF AZTHREONAM ADMINISTERED AS A 30-MIN INTRAVENOUS INFUSION (Assay by Squibb Institute)

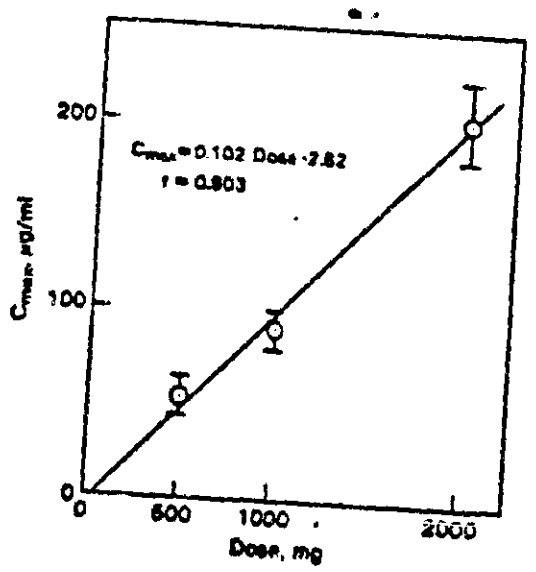
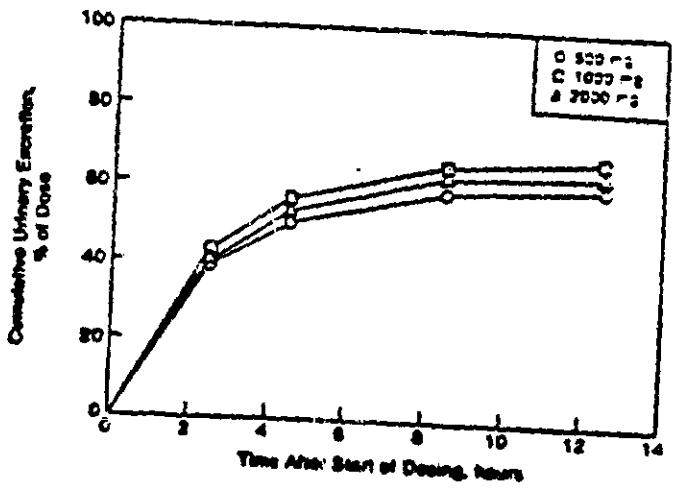


FIGURE 4

CUMULATIVE URINARY EXCRETION OF AZTHREONAM BY BIOASSAY UP TO 12.5 HR AFTER THE START OF 30-MIN INTRAVENOUS INFUSIONS OF AZTHREONAM (Assay by Squibb Institute)



G. Study Protocol #18,554-8 (Pivotal Study):

1. Title: Intravenous safety and pharmacokinetic study of aztreonam in patients with renal insufficiency.

2. Objective: The objectives of this study were to obtain safety and pharmacokinetic data on aztreonam in patients with renal dysfunction and healthy control subjects.

3. Study Design: Enrolled in this study were 24 male volunteers between the ages of 24 and 64 years (body weights ranges between 63 and 97 kg). The volunteers were assigned to study groups based upon their urinary creatinine clearance determined at the time of screening. Excluded were volunteers who has abnormal hepatic function (i.e., abnormal SGOT, SGPT or total serum bilinubin).

Each volunteer received a single 1000 mg IV dose of aztreonam given as a 2 minute infusion. Prior to drug administration patients were fasted for 8 and then for 2 hours post-drug administration. 250 ml of water were ingested at the time of dosing and then at 1 and 2 hours post-dosing. Beverages not excluded (i.e. caffeine-containing) were permitted ad libitum.

Drug was supplied as a sterile powder blend of aztreonam, and L-arginine (ratio=1.0/0.7) for reconstintion in sterile water.

Serum and urine samples were serially collected over 48 hours post-drug administration. Extra serum samples were also collected at 0, 10 min, 1 and 3 hours in order to determine drug serum protein binding. Drug concentrations in serum, protein-free filtrate and urine were determined by the microbiological agar diffusion assay. Concentrations of aztreonam, as well as the metabolite SQ 26,992, in serum and urine were also determined by a HPLC assay method. The sponsor indicated that results for aztreonam in serum and urine were equivalent as determined by both the bioassay and HPLC methods. However, because the bioassay had lower quantitation limits, results for that method were provided.

Pharmacokinetic analyses were done using a open two compartment model approach. The analytical methodology used for assessing aztreonam's recommended dose adjustments in renal impairment are found in Attachment II.

The clinical portions of the study were conducted by Drs. W.K. Bolton and W.M. Schell, University of Virginia, School of Medicine, Charlotesville, VA. The sample analyses were conducted by the Squibb Insititute.

4. Results:

a. Table 1 provides information on the group assignments of study patients as related to pre-dose creatinine clearance. Table 3 gives the concomitant medications for those patients tested in this study.

b. Table 38 gives mean aztreonam serum concentrations for each study group and Table 41 gives mean urinary drug concentration results.

c. Table 40 gives individual subjects' AUC_{0-48} values and Table 40A gives summary pharmacokinetic data results.

d. Table 56 gives correlation results of different pharmacokinetic parameters with urinary creatinine clearance. Tables 57 and 59 give recommended dosing adjustments for renal dysfunction using dose and dosage interval modifications, respectively. Table 58 gives the PK parameters used to predict drug serum levels using the different dosing adjustments (Figures 10-14).

e. Figure 1 gives mean aztreonam serum concentrations for each study group. Figures 2 and 4 give the correlation plots for serum and urine drug concentrations determined by both the bioassay and HPLC methods.

f. Figure 3 gives the percent urinary excretion of aztreonam for the different study groups with different degrees of renal dysfunction.

g. Figures 7 and 8 give serum and renal clearance correlations with urinary creatinine clearance, respectively.

h. Figures 10-14 give "predicted" aztreonam serum levels for patients with different degrees of renal insufficienty using either adjusted doses or adjusted dosing intervals.

5. Comments:

a. Attachment I gives the sponsor's summary of the study's pharmacokinetic findings. These appear to be accurate.

b. In the proposed package insert in the Dosage and Recommendation section the sponsor has provided an equation to estimate renal creatinine clearance when only a serum creatinine concentration is available in patients with renal insufficiency. Using this calculated creatinine clearance, dose adjustment is then recommended if appropriate. The sponsor has however recommended not to use serum creatinine levels to calculate renal creatinine clearance in elderly patients. Instead renal creatinine clearance should be determined before making dose adjustments. This is appropriate since in the elderly a general decrease in renal function is normally observed but serum creatinine concentration remains remarkly constant.

6. Conclusion: Study #18,554-8 is an acceptable study in that it demonstrated the effects of decreased renal function on aztreonam's pharmacokinetics following a single 1000 mg 2 minute intravenous dose given to paitents with different degrees of renal impairment. Based upon the study's findings the investigators have recommended aztreonam dose adjustments for different degrees of renal failure. Section IV of this review should be further reviewed regarding this study's findings.

Group No.	Pre-Dose Creatinine Clearance, ml/min	No. of Volunteers	Patient Nos.
I	>80	8	1-6,8,12
II	30-80	5	7,9,11,13,15
III	10-29	5	10,14,16-18
IV	<10	6	19-24

TABLE 1

Protocol 18334-8
CONCOMITANT MEDICATIONS

GROUP: 1

PATIENT NUMBER	CONCOMITANT MEDICATION	TOTAL DAILY DOSE	UNITS
06	Propranolol	800	MG
	Hydralazine	100	MG
	Hydrochlorothiazide	100	MG
	Ampicillin	650	MG
12	Prednisolone	10	MG
	Cimetidine	800	MG

GROUP: 2

PATIENT NUMBER	CONCOMITANT MEDICATION	TOTAL DAILY DOSE	UNITS
07	Propranolol	320	MG
	Allopurinol	300	MG
	Methyldopa	2000	MG
	Chlorthalidone	100	MG
	KCL	20	MEQ
09	Prednisolone	10	MG
	Furosemide	200	MG
	Hydralazine	75	MG
	Aminophylline	800	MG
	Colchicine	2	MG
	Tetracycline Sulfate	*	
11	Hydrochlorothiazide	100	MG
	Allopurinol	300	MG
	Acetaminophen	*	
13	Prednisolone	45	MG
	Propranolol	240	MG
	Furosemide	80	MG
	Hydralazine	150	MG
15	Methyldopa	750	MG
	Hydrochlorothiazide	50	MG

*Those not recorded, or administered only as needed (PRN).

GROUP: 3

PATIENT NUMBER	CONCOMITANT MEDICATION	TOTAL DAILY DOSE	UNITS
14	Insulin Isophane	40	U
	Diazepam	10	MG
	Hydrochlorothiazide	50	MG
	Vitamin C	500	MG
	Cimetidine	600	MG
	Diphenhydramine HCL	200	MG
	Folic Acid	1	MG
	Ferrous Gluconate	325	MG
	Quinine Sulfate	324	MG
16	Prednisolone	5	MG
	Acetaminophen	3000	MG
	Mefenamic Acid	150	MG
17	Insulin Isophane	15	U
	Hydrochlorothiazide	100	MG

GROUP: 4

PATIENT NUMBER	CONCOMITANT MEDICATION	TOTAL DAILY DOSE	UNITS
19	Valproate Na	750	MG
	Cimetidine	600	MG
	Ca Carbonate	6300	MG
	Ferrous Fumarate	1	MG
	Cholecalciferol	1	MG
	Chloral Hydrate	*	
	Aluminum Hydroxide	11	G
20	Ca Carbonate	3900	MG
	Propoxyphene HCL	*	
	Ferrous Sulfate	600	MG
	Ferrous Fumarate	1	MG
	Aluminum Hydroxide	4	G
	Flurazepam HCL	*	
21	Ca Carbonate	1300	MG
	Ferrous Fumarate	1	MG
	Cholecalciferol	1	MG
	Aluminum Hydroxide	7300	MG
22	Propranolol	160	MG
	Aluminum Hydroxide Mg	3	G
	Ferrous Fumarate	1	MG

SERUM CONCENTRATIONS (MEAN ± SEM, uGM/ML) OF AZTREONAM AFTER A SINGLE INTRAVENOUS DOSE OF 1000 MG USING BIOASSAY

TIME AFTER INFUSION HR	GROUP I N=8	GROUP II N=5	GROUP III N=5	GROUP IV N=6
Pre	0.0	0.0	0.0	0.0
0.17	81.2 ± 3.7	98.4 ± 4.6	101.2 ± 12.0	145.1 ± 27.0
0.33	64.2 ± 2.0	75.7 ± 0.9	78.4 ± 6.5	74.9 ± 10.1
0.50	55.2 ± 2.2	68.8 ± 1.3	68.1 ± 5.5	67.6 ± 7.4
1.0	39.6 ± 1.7	55.4 ± 1.4	58.5 ± 5.5	56.6 ± 3.7
2.0	27.0 ± 1.6	44.6 ± 2.4	46.6 ± 4.1	46.7 ± 1.8
3.0	17.9 ± 1.2	35.2 ± 2.9	39.2 ± 2.6	40.8 ± 1.4
4.0	12.6 ± 1.2	29.1 ± 2.8	33.7 ± 3.0	35.0 ± 1.2
6.0	6.1 ± 0.9	19.5 ± 2.7	25.3 ± 2.6	27.7 ± 2.2
8.0	3.4 ± 0.7	14.0 ± 2.6	18.7 ± 2.0	21.8 ± 1.8
12.0	0.93 ± 0.24	8.4 ± 1.4[a]	11.3 ± 1.5	13.9 ± 1.7
24.0	0.01 ± 0.01	0.84 ± 0.27	2.0 ± 0.3	4.4 ± 1.4
48.0	0.0	0.0	0.05 ± 0.02	0.48 ± 0.2

[a]Omitted 12-hr value of 155.0 ug/ml for patient #7.

TABLE 41

URINARY CONCENTRATIONS (MEAN ± SEM, uGM/ML) OF AZTREONAM AFTER A SINGLE INTRAVENOUS DOSE OF 1000 MG USING BIOASSAY

TIME AFTER INFUSION HR	GROUP I N=8	GROUP II N=5	GROUP III N=5	GROUP N=2
0-2	2161 ± 489	516 ± 147	449 ± 134	61.5 ± 39
2-4	645 ± 201	486 ± 294	365 ± 212	45.7 ± 30
4-8	213 ± 46	201 ± 72	200 ± 50	80.5 ± 0
8-12	59.4 ± 11.6	94.5 ± 30.0	111 ± 34	63.4 ± 19
12-24	9.16 ± 1.14	16.8 ± 2.9	31.9 ± 7.5	93.3 ± 35
24-48	0.18 ± 0.04	1.56 ± 0.34	2.8 ± 0.7	5.6 ± 4

[a]Data are summarized for two patients who provided urine samples for all collection periods.

TABLE 40

AREA (TRAPEZOIDAL RULE) UNDER SERUM CONCENTRATION-TIME CURVE, $AUC_{0-48\ hr}$ (μGM x HR/ML), OF ACTINOMYCIN AFTER A SINGLE INTRAVENOUS DOSE OF 1600 μG/M² DURING RADIATION

CrCl > 80 | 70-80 | 10-29 | < 10

GROUP I		GROUP II		GROUP III		GROUP IV	
PATIENT NO.	$AUC_{0-48\ hr}$	PATIENT NO.	$AUC_{0-48\ hr}$	PATIENT NO.	$AUC_{0-48\ hr}$	PATIENT NO.	$AUC_{0-48\ hr}$
1		7		10		19	
2		9		14		20	
3		11		16		21	
4		13		17		22	
5		15		18		23	
6						24	
8							
12							
MEAN ± SEM	171.3 ± 12.0		377.8 ± 33.6		475.1 ± 40.1		809.3 ± 192.0

Table 40 A
Summary Pharmacokinetic Results
(Mean (CV) Range)

Parameter	Cl_{CR} >80 (ml/min) I	30–80 II	10–29 III	<10 IV
	Groups			
$t_{1/2\alpha}$ (hrs)	0.22(39, 0.12-0.4)	0.31(87, 0.1-0.8)	0.21(43, 0.1-0.34)	0.13(75, 0.02-(
$t_{1/2\beta}$ (hrs)	1.94(20, 1.56-2.57)	3.67(23, 2.74-4.53)	4.73(20, 3.3-5.59)	6.02(33, 4.59-(
K_{12} (hr^{-1})	1.39(69, 0.33-3.31)	1.75(100, 0.14-4.63)	2.01(75, 0.62-4.33)	5.04(91, 1.06-
K_{21} (hr^{-1})	1.82(22, 1.23-2.17)	1.62(36, 0.68-2.21)	1.69(22, 1.38-2.27)	1.37(77, 0.22
K_{10} (hr^{-1})	0.69(20, 0.45-0.89)	0.39(29, 0.20-0.50)	0.38(76, 0.18-0.9)	3.96(180, 0.2-(
			n=4	0.30(7, 0.2-0.3
V_1 (L/Kg)	0.12(29, 0.06-0.13)	0.10(45, 0.05-0.19)	0.08(28, 0.04-0.11)	0.06(82, 0.004-0
V_{ss} (L/Kg)	0.2(14, 0.16-0.22)	0.17(26, 0.14-0.23)	0.16(14, 0.14-0.2)	0.16(46, 0.02-(
V_{darea} (L/Kg)	0.22(13, 0.18-0.25)	0.18(25, 0.15-0.24)	0.17(13, 0.15-0.20)	0.18(27, 0.08-
Serum Protein Binding (%)	53.6(12, 40.1-58)	44.2(22, 22.8-53.3)	46.4(9, 41.7-49.4)	40.0(19, 31.3-
Urinary Exc (%) 0-48	56.1(12, 47.9-67.8)	31.2(25, 21.7-40.8)	22.2(52, 13.3-42.4)	1.4(175, 0-
Cl_R (ml/min)	14.3(18, 7.8-17.4)	48.7(25, 36.3-64.8)	28.3(20, 20.9-29.8)	7.5(37#, 1
Cl_R (ml/min)	57.2(24, 35.8-76.6)	15.9(48, 7.9-26.4)	8.1(41, 5.9-14)	0.5(196, 0-

TABLE 16

DEPENDENCE OF PHARMACOKINETIC PARAMETERS FOR AZITHROMYCIN ON URINARY CREATININE CLEARANCE

REGRESSION EQUATION	CORRELATION COEFFICIENT, r	P-VALUE FOR SLOPE
$C_{max} = 126 - 0.403\ Cl_{Cr}$	0.519	0.0093
$AUC = 665 - 4.13\ Cl_{Cr}$	0.668	0.0004
$t_{½\alpha} = 0.185 + 0.00059\ Cl_{Cr}$	0.203	0.341
$k_{12} = 3.74 - 0.0224\ Cl_{Cr}$	0.428	0.0367
$k_{21} = 1.51 + 0.00234\ Cl_{Cr}$	0.198	0.353
$V_1 = 0.0648 + 0.00050\ Cl_{Cr}$	0.603	0.0018
$V_{ss} = 0.153 + 0.00035\ Cl_{Cr}$	0.447	0.0284
$V_{area} = 0.168 + 0.00079\ Cl_{Cr}$	0.539	0.0065
$\%\ bound = 41.0 + 0.104\ Cl_{Cr}$	0.661	0.0004
$t_{½\beta} = 5.51 - 0.0290\ Cl_{Cr}$	0.806	0.0001[a]
$k_{10} = 2.21 - 0.0151\ Cl_{Cr}$	0.225	0.291
urinary excr. Az. $\%\ dose\ Az. = 8.88 + 0.386\ Cl_{Cr}$	0.925	0.0001[b]
urinary excr. SQ 26,992. $\%\ dose\ Az. = 4.48 + 0.0306\ Cl_{Cr}$	0.341	0.103
$Cl_{Az,s} = 25.2 + 0.609\ Cl_{Cr}$	0.968	0.0001
$Cl_{Az,r} = -1.76 + 0.470\ Cl_{Cr}$	0.989	0.0001
$Cl_{Az,ur} = 26.9 + 0.138\ Cl_{Cr}$	0.704	0.0001

[a] The relationship of $t_{½\beta}$ vs. Cl_{Cr} was nonlinear (Figure 5), although approximately 55% of the variance ($r^2 = 0.65$) could be related to the linear regression line.

[b] The relationship of urinary excr. Az. vs Cl_{Cr} was nonlinear (Figure 6), although approximately 86% of the variance ($r^2 = 0.86$) could be related to the linear regression line.

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TABLE 31

DOSAGE REDUCTION FOR AZTREONAM ADMINISTERED TO PATIENTS WITH RENAL INSUFFICIENCY: CONSTANT DOSAGE INTERVAL, VARIABLE DOSE

PATIENT CREATININE CLEARANCE ML/MIN	DOSE REDUCTION FACTOR Cl_R/Cl_N	CATEGORY OF RENAL INSUFFICIENCY	CATEGORY OF DOSAGE, FRACTION OF NORMAL[a]
125.0 81	1 0.76	NORMAL	1
80 55 30	0.73 0.58 0.43	MILD	1/2[b]
29 20 10	0.42 0.37 0.31	MODERATE	1/3[b]
9 5 1 0	0.30 0.28 0.24 0.23	SEVERE	1/4[b]

[a] Designed to maintain approximately constant mean serum level of aztreonam for various degrees of renal insufficiency, assuming the dose interval is the same for all stages of renal disease. For example, if the standard dose is 1000 mg q8h in patients with normal renal function, then a patient with a creatinine clearance less than or equal to 9 ml/min would receive 250 mg q8h.

[b] All patients with renal insufficiency should receive a loading dose equal to the dose used in patients with normal renal function.

TABLE 30

PREDICTED PHARMACOKINETIC PARAMETER VALUES FOR REPRESENTATIVE CREATININE CLEARANCES

Group	Range of Cl_{Cr} ml/min	Representative Cl_{Cr}, ml/min	Predicted Kinetic Parameter Values[a] α, hr^{-1}	β, hr^{-1}	k_{21}, hr^{-1}	V_1, liters
I	>80	124	3.25	0.363	1.75	9.42
II	30-80	55	3.82	0.226	1.16	8.04
III	10-29	20	4.10	0.156	1.77	7.34
IV	<10	5	4.22	0.126	1.77	7.04

[a] See Appendix D

TABLE 32

PROPOSED DOSAGE DOSE INTERVAL PROLONGATION FOR AZTREONAM ADMINISTERED TO PATIENTS WITH RENAL INSUFFICIENCY: CONSTANT DOSE, VARIABLE DOSE INTERVAL

PATIENT CREATININE CLEARANCE ML/MIN	DOSE INTERVAL PROLONGATION FACTOR Cl_R/Cl_N	CATEGORY OF RENAL INSUFFICIENCY	DOSAGE INTERVAL, MULTIPLE OF NORMAL[a]
125.0 81	1 0.35	NORMAL	1
80 55 30	1.37 1.72 2.32	MILD	2
29 20 10	2.38 2.70 3.22	MODERATE	3
9 5 1 0	3.33 3.57 3.85 4.00	SEVERE	4

[a] Designed to maintain approximately constant mean serum level of aztreonam for various degrees of renal insufficiency, assuming the dose is the same for all stages of renal disease. For example, if the standard dose is 1000 mg q8h in patients with normal renal function, then a patient with a creatinine clearance less than or equal to 9 ml/min would receive 1000 mg q12h.

FIGURE 7

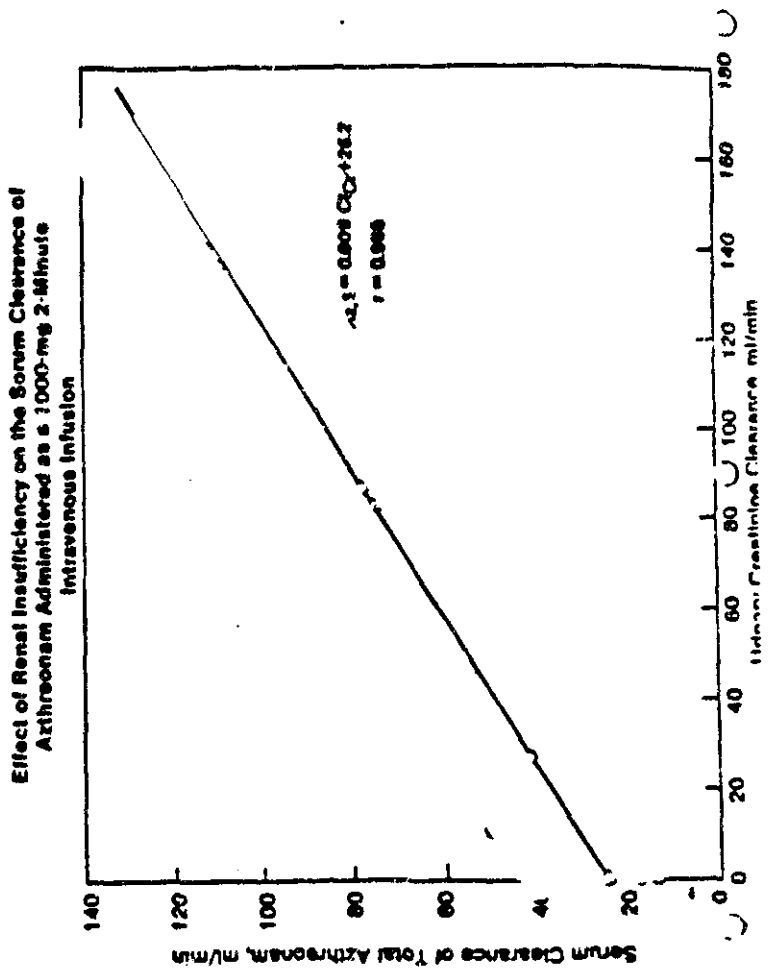
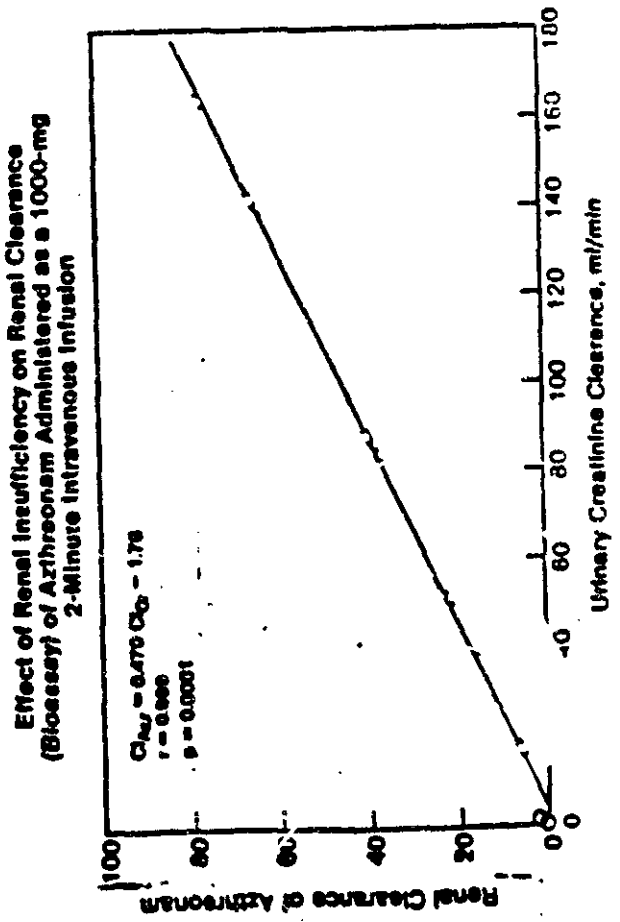


FIGURE 8



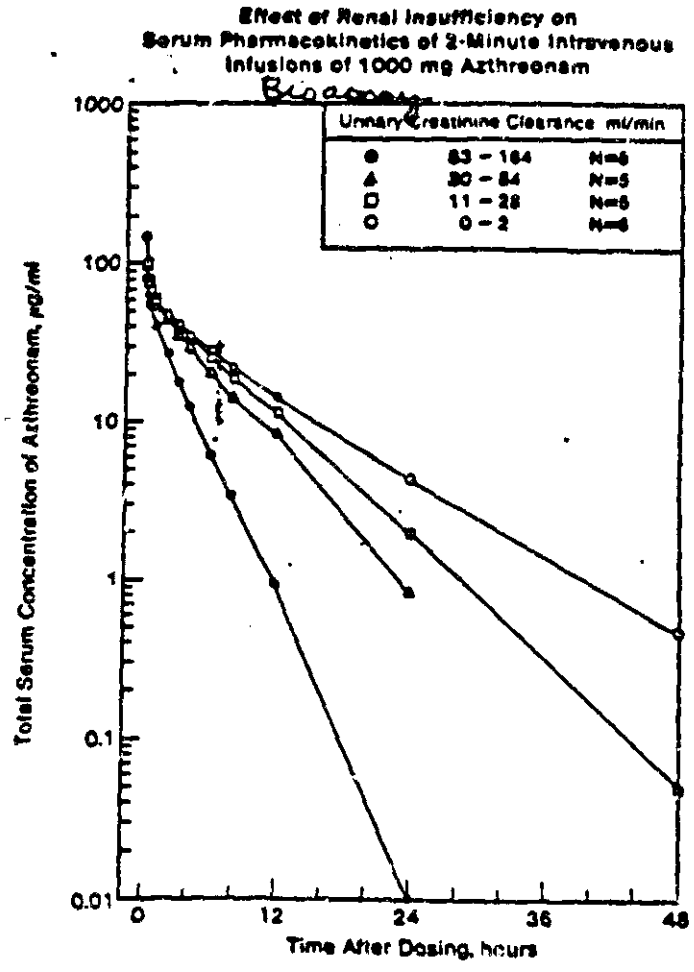


FIGURE 2

FIGURE 3

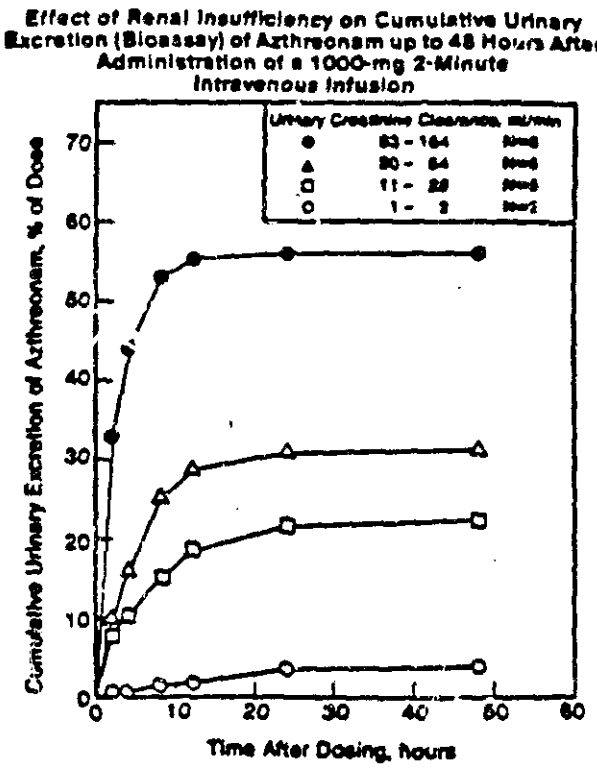
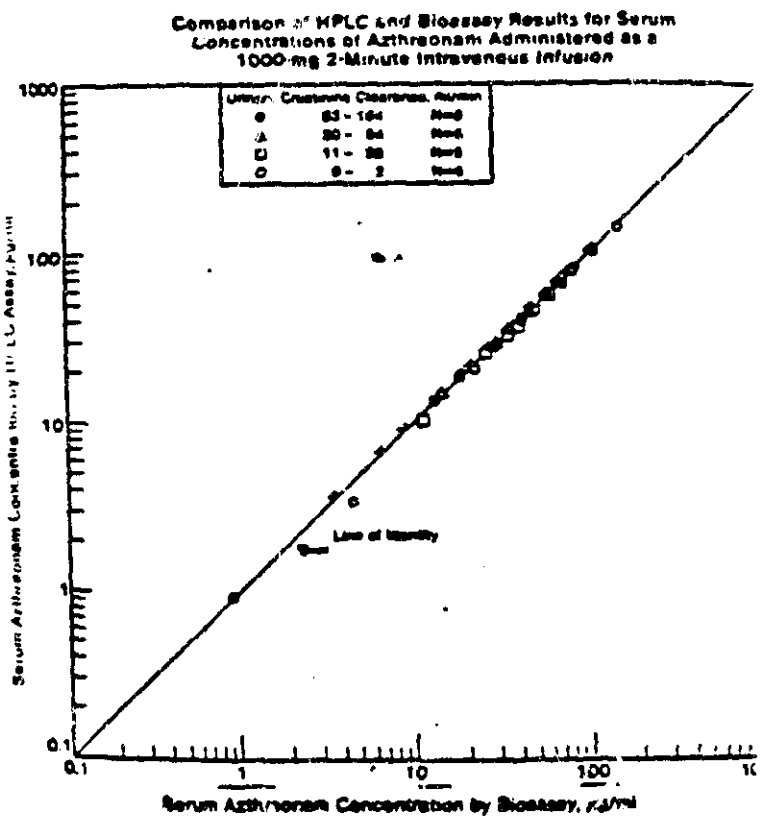


FIGURE 4



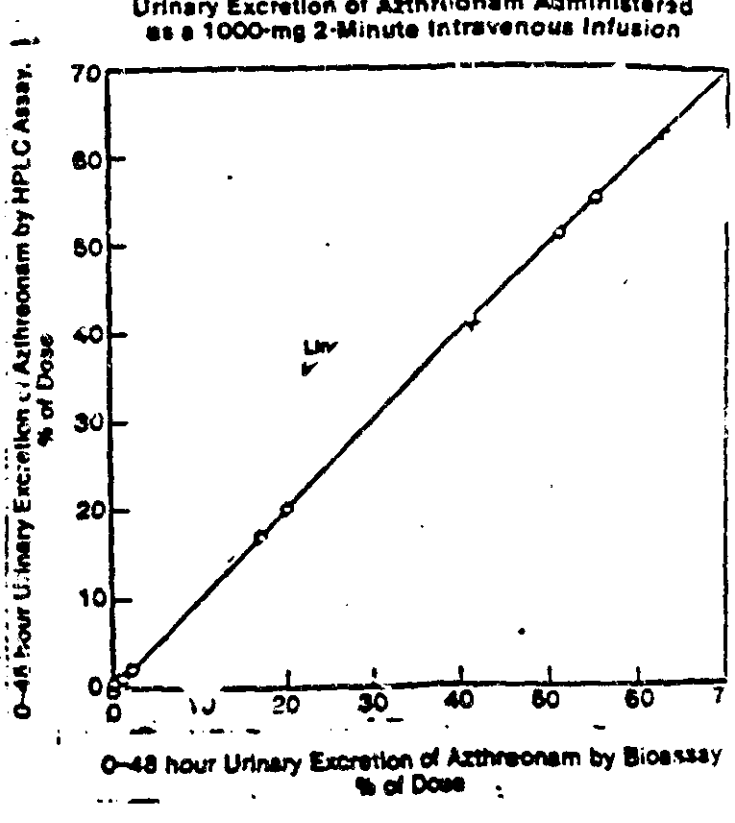


FIGURE 10

Predicted Serum Azthreonam Concentrations for a Dosage Regimen Modified for Renal Insufficiency: Constant Dosage Interval (8 Hours) and Variable Dose (Beginning at 1000 mg)

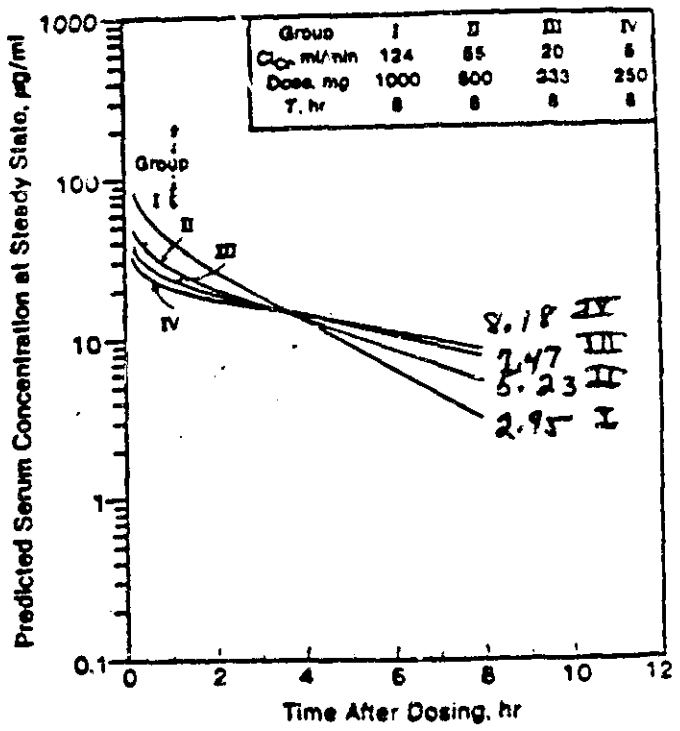


FIGURE 11

Predicted Serum Azthreonam Concentrations for a Dosage Regimen Modified for Renal Insufficiency: Constant Dose (1000 mg) and Variable Dosage Interval (Beginning at 8 Hours)

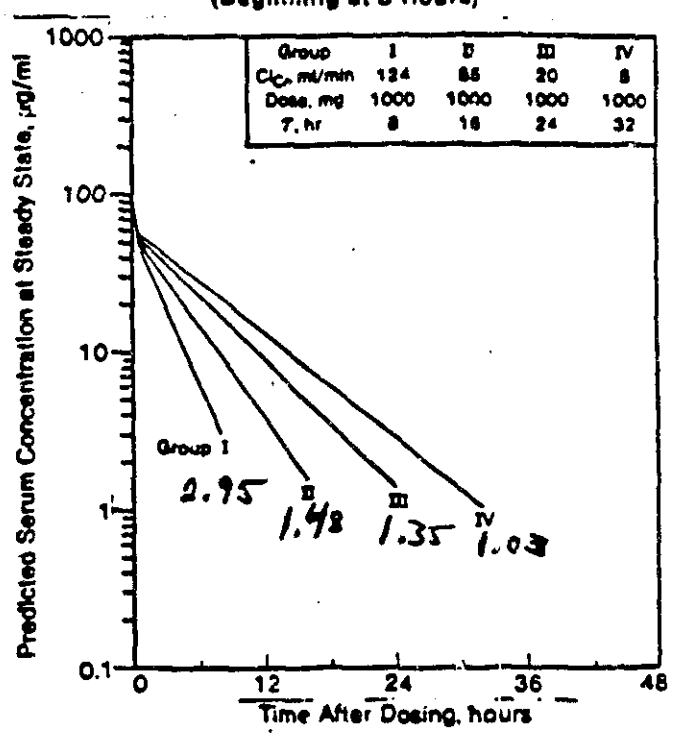
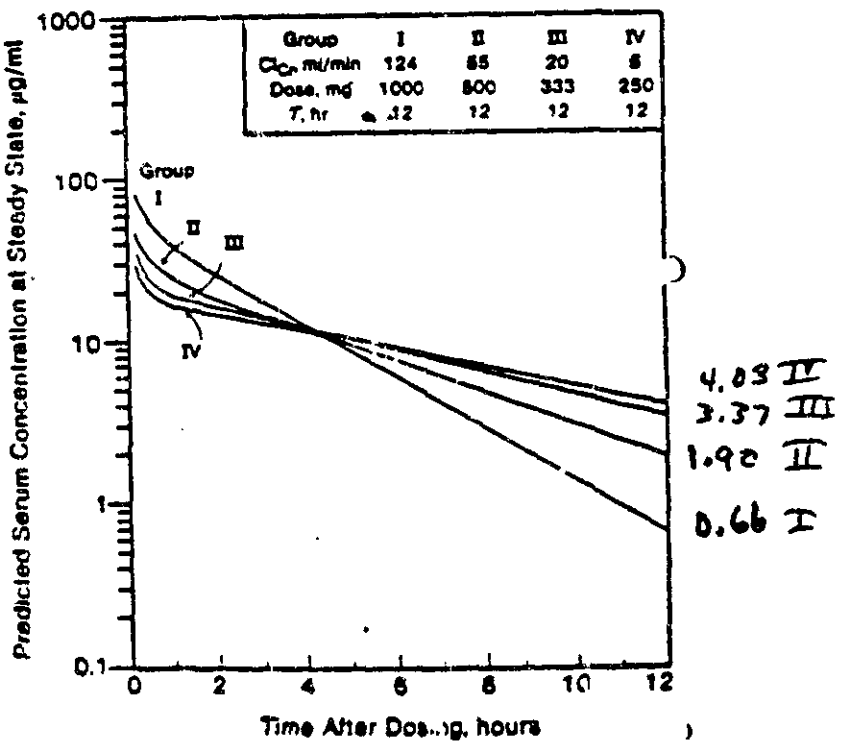


FIGURE 12

Predicted Serum Azthreonam Concentrations for a Dosage Regimen Modified for Renal Insufficiency: Constant Dosage Interval (12 Hours) and Variable Dose (Beginning at 1000 mg)



Predicted Serum Azthreonam Concentrations for a Dosage Regimen Modified for Renal Insufficiency: Constant Dose (1000 mg) and Variable Dosage Interval (Beginning at 12 Hours)

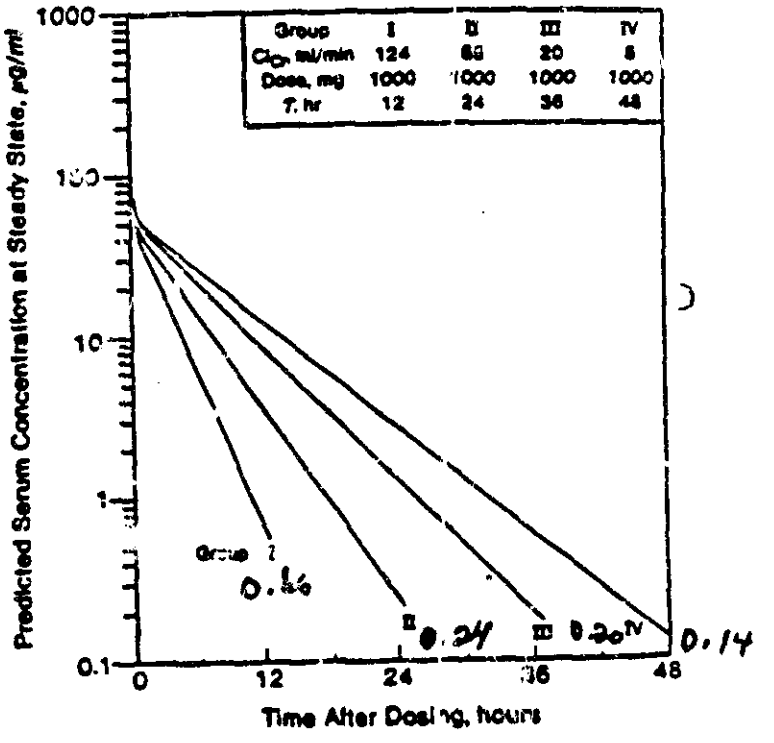


Table I. Pharmacokinetic Analysis of Aztreonam Data

mean (S.E.M.)

Parameter	Biliary Cirrhosis	Alcoholic Cirrhosis	Normal Subjects
C_{max}, ug/ml	103.20 ± 13.51	115.40 ± 16.43	114.40 ± 14.49
$AUC_{0-12\ hr}$ ugxhr/ml	237.60 ± 21.16	231.10 ± 21.10	189.40 ± 15.00
Distribution			
*Extent			
V_1, liters/kg	0.12 ± 0.02	0.08 ± 0.02	0.06 ± 0.02
V_{DSS}, liters/kg	0.18 ± 0.02	0.18 ± 0.02	0.15 ± 0.02
$V_{D\ AREA}$, liters/kg	0.19 ± 0.02	0.22 ± 0.03	0.17 ± 0.01
serum protein binding %	69.62 ± 1.36	69.13 ± 1.72	73.02 ± 2.02
*Rate			
$t_{\frac{1}{2}\alpha}$, hr_{-1}	0.28 ± 0.09	0.36 ± 0.17	0.14 ± 0.04
k_{12}, hr_{-1}	1.82 ± 0.72	3.21 ± 1.31	3.61 ± 1.04
k_{21}, hr^{-1}	2.16 ± 0.53	1.12 ± 0.27	1.87 ± 0.27
Elimination			
*Extent			
12-hr urinary excr. % of Dose	54.41 ± 6.73	*75.53 ± 7.22	62.41 ± 5.55
serum clearance, ml/min/kg	1.00 ± 0.08	***0.82 ± 0.04	1.08 ± 0.12
renal clearance, ml/min/kg	0.55 ± 0.10	0.63 ± 0.08	0.69 ± 0.11
nonrenal clearance, ml/min/kg	**0.45 ± 0.07	0.19 ± 0.05	**0.39 ± 0.01
*Rate			
$t_{\frac{1}{2}\beta}$, hr_{-1}	***2.17 ± 0.06	***3.24 ± 0.56	1.89 ± 0.17
k_{10}, hr^{-1}	0.61 ± 0.10	1.47 ± 0.78	2.10 ± 1.13

*Significantly different from biliary cirrhosis mean ($P<0.05$).
**Significantly different from alcoholic cirrhosis mean ($P<0.01$ and $P<0.05$ for biliary cirrhosis and healthy subjects respectively).
***Significantly different from the mean for healthy subjects ($P<0.05$).

TABLE 6

MEAN CUMULATIVE URINARY EXCRETION OF SO 26,992
as Measured by HPLC
(in mg)

Biliary Alcoholic Normal

0 to 12 Hrs	Mean:	14.482	15.760	19.028
	SEM:	(2.4036)	(1.5640)	(3.3327)
	n:	5	6	6
	% Dose	1.5%	1.6%	1.9%

Figure 1
Effect of Hepatic Disease on Serum Pharmacokinetics of 3-Minute Intravenous Infusions of 1 gm Aztreonam

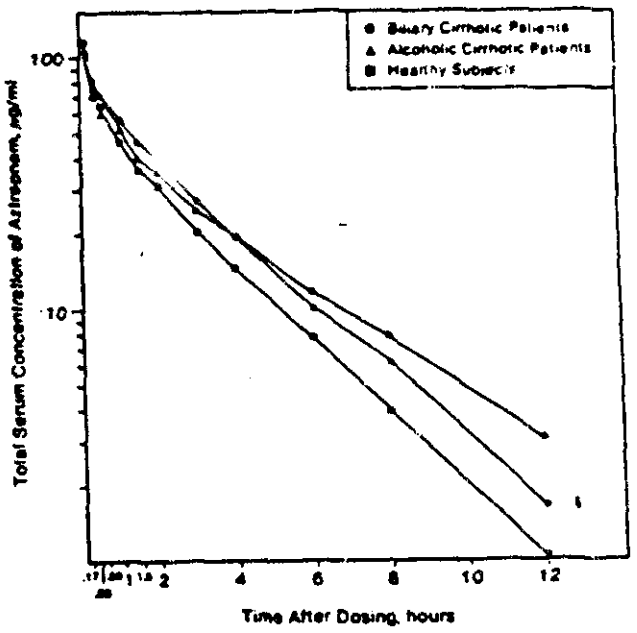
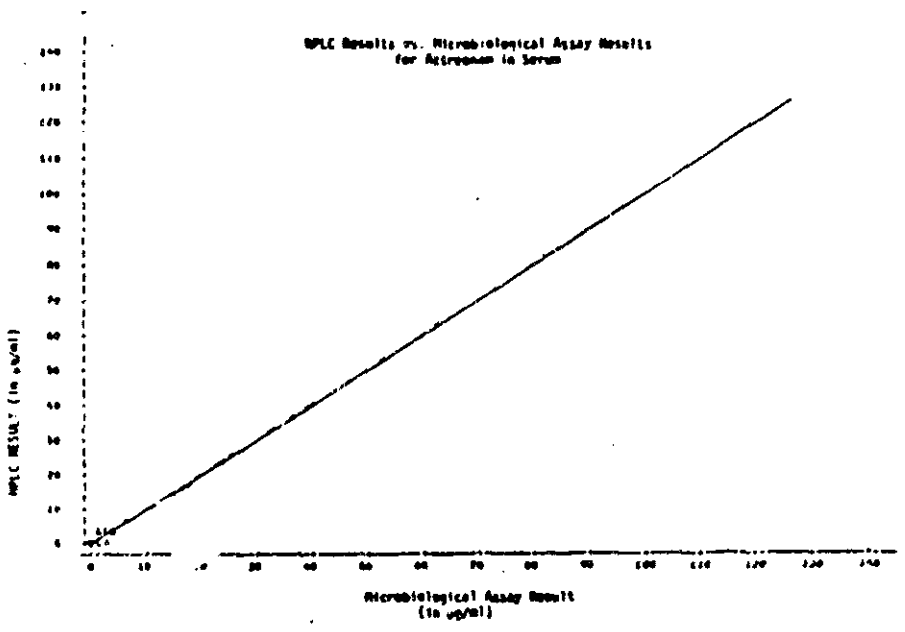


TABLE 1A

SUMMARY OF
SERUM CONCENTRATION OF AZTREONAM
As measured by Microbiological Assay
(Means and S.E.M.'s)
(in ug/ml)

Time After Infusion (in hours) n=	Biliary Cirrhosis Patients 2	Alcoholic Cirrhosis Patients 6	Healthy Subjects 6
Prior to Infusion	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
0.17	103.2 (13.5)	109.8 (17.7)	114.4 (14.5)
0.33	82.5 (11.0)	77.9 (9.6)	70.9 (2.8)
0.50	72.2 (7.2)	60.1 (4.6)	64.1 (2.9)
1.00	57.5 (5.5)	55.1 (7.1)	46.1 (3.3)
1.50	46.7 (3.7)	40.0 (3.2)	36.7 (2.2)
2.0	40.7 (3.1)	36.0 (3.1)	31.2 (2.5)
3.0	27.3 (2.4)	25.2 (2.3)	20.6 (1.8)
4.0	19.9 (1.7)	19.6 (2.2)	14.3 (1.1)
6.0	10.7 (1.1)	11.6 (1.4)	7.6 (1.4)
8.0	6.1 (0.6)	7.6 (1.0)	3.9 (0.9)



Solid line represents HPLC = Micro.

"A" represents 1 observation. "B" represents 2 observations, etc.

VII. ADVERSE REACTIONS

Four of the 18 volunteers experienced adverse reactions during the study. Patient 1 (a biliary cirrhotic patient) had moderate abdominal pain 1 hour after eating on the same day aztreonam was administered. The pain subsided within 30 minutes without treatment and was considered by the investigator to be related to the patients' cholecystectomy. Subject 13 (a healthy subject) had mild abdominal discomfort after dinner 10 hours following aztreonam administration. The discomfort lasted 1 hour and disappeared without treatment. This was also not considered to be related to aztreonam by the investigator. Subject 15 (a healthy subject) experienced mild fatigue and mild difficulty in concentrating on Day 3. Both of these effects were considered to be work related. Subject 17 (a healthy subject) had several loose stools for 6 hours beginning 14 hours after drug infusion. The condition subsided without treatment and was considered to be possibly related to

Attachment I

Mezlocillin kinetics are altered in hepatic disease (Sunke et al, 1983). In patients with alcoholic cirrhosis, the terminal half-life of mezlocillin was almost three times longer than that in healthy subjects and nonrenal clearance was markedly reduced (by 90%). The authors recommended dosage reduction of mezlocillin in hepatic patients according to the following equation:

$$F_p = \frac{AUC_N}{AUC_p}$$

where F_p is the dose fraction of a drug for a given patient with decreased clearance of that drug, AUC_p is the AUC for that patient and AUC_N is the AUC for patients with normal clearance of the drug. The dose fraction in that study for patients with hepatic disease was 0.51. The dose fraction is multiplied by the normal dose to obtain the reduced dose for cirrhotic patients. Thus the patients in that study would receive only half the usual dose. This method of calculating dosage reduction assures that the AUC in patients with reduced clearance will remain constant and equal to that in normal patients. If, on the basis of clinical status or anticipated duration of therapy, dosage reduction of aztreonam becomes desirable, dosage could be reduced according to the following formula (AUCs from Table 6):

$$F_p = \frac{AUC_N}{AUC_p} = \frac{189}{231} = 0.82$$

Thus, for alcoholic cirrhotics, the dose would be reduced by 18%.

Another method for calculating dosage reduction is based on comparison of serum clearances, i.e., the dose fraction is derived by dividing the serum clearance in patients by the serum clearance in normals (Aronoff et al, 1981). For the present study (serum clearances from Table 8) the dose fraction becomes: 0.82 ÷ 1.08 = 0.76, and the dose for alcoholic cirrhotics would be 76% of the normal dose. In practice, the physician, who was concerned about the dose of aztreonam in an alcoholic cirrhotic patient, could reduce the dose by 20-25% and be reasonably sure that the AUC in that patient would be similar to that in patients with normal clearances.

DEPARTMENT
Department of Clinical Pharmacology

SECTION
Division of Medical Affairs

DATE OF REPORT
April 5, 1993

NUMBER
HMR-860

PRODUCT NAME OR GENERIC NAME
SQ 26,776 (Aztreonam)

TITLE
Report on Comparison of Safety, Pharmacokinetics, and Serum and Urine Bactericidal Activity of Intravenous Aztreonam and Moxalactam in Healthy Subjects. Study Protocol # 18554-23

AUTHORS
Edward A. Swabb, M.D., Ph.D., May Frantz, Ph.B., and Tricia Yeh, M.S.

Lilly

Reed Luthy, M.D., Department of Medicine, University of Zurich

ABSTRACT

Aztreonam and moxalactam were each administered as a single 2000-mg intravenous infusion over 30 minutes to 6 healthy male volunteers according to a two-way crossover study design with a 7-day washout period between drug treatments. To assess the safety of the drug treatments, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and at frequent intervals after each drug treatment.

Aztreonam and moxalactam were tolerated well by 6 healthy male subjects. Possible drug-related adverse reactions after administration of aztreonam consisted of mild diaphoresis (1 subject) and mild fatigue (2 subjects), and after administration of moxalactam consisted of mild diarrhea and dizziness (1 subject) and mild diarrhea and flatulence (1 subject). These findings were reversible without specific treatment.

The pharmacokinetic profiles of aztreonam and moxalactam were assessed by measuring aztreonam and moxalactam (sum of R and S epimers) concentrations in multiple samples of serum and urine after administration of the antibiotics. Assays were performed by the clinical investigator using a high-pressure liquid chromatography method. Mean values for the concentrations of aztreonam and moxalactam in serum and urine are shown in Table I.

Table I

Time After Start of Infusion, hr	Serum[c] Aztreonam, ug/ml	Serum[c] Moxalactam, ug/ml
Pre	0 ± 0	0 ± 0
0.5[a]	137.1 ± 2.3	169.2 ± 3.3
2	51.8 ± 1.7	63.0 ± 1.7
6	13.3 ± 0.6	18.0 ± 0.9
8	6.7 ± 0.2	8.6 ± 0.3
12	1.8 ± 0.2	2.2 ± 0.1
Time After Start of Infusion, hr	**Urine[c] Aztreonam, ug/ml**	**Urine[c] Moxalactam, ug/ml**
Pre	0 ± 0	8 ± 0
0-0.5[b]	6120 ± 1367	6534 ± 1203
0.5-2	6010 ± 1349	6479 ± 1652
2-6	2139 ± 573	2977 ± 754
6-8	957 ± 278	1475 ± 121
8-12	311 ± 43	735 ± 120

[a]At the end of the 30-minute infusion.

[b]Collected during the 30-minute infusion.

[c]Values are arithmetic mean ± SEM for 6 subjects.

Maximum serum concentrations (C_{max}), areas under the serum concentration-time curve (AUC), elimination half-life ($t_{1/2}$), and urinary recovery are shown in Table II. Although moxalactam gave statistically significantly greater mean values for C_{max} and AUC, none of the differences shown in Table II was considered to be of therapeutic importance.

Table II

Parameter[a]	Aztreonam	Moxalactam	p[b]
C_{max}, ug/ml	137.1 ± 2.3	169.2 ± 3.3	<0.01
AUC_{0-12hr}, ug × hr/ml	343.1 ± 5.5	426.8 ± 7.1	<0.01
$t_{1/2}$, hr	2.07 ± 0.11	2.01 ± 0.07	NS
Urinary recovery, % of dose, 0-8 hr	58.5 ± 5.4	64.8 ± 4.3	NS

[a]Values are arithmetic mean ± SEM for 6 subjects.

[b]Based upon analysis of variance for the crossover design.

The pharmacokinetics of aztreonam described in this study were consistent with previously reported results for 30-minute intravenous infusions of a 2-gram dose in healthy volunteers (Protocol 18554-18).

Serum and urinary bactericidal titers were determined by the clinical investigator at the same times as shown in Table I for the six test organisms shown in Table III.

Table III

Bacterial Strain	Aztreonam MIC, ug/ml	Aztreonam MBC, ug/ml	Moxalactam MIC, ug/ml	Moxalactam MBC, ug/ml
Escherichia coli	0.06	0.125	0.125	0.25
Klebsiella pneumoniae	0.06	0.125	0.125	0.5
Proteus mirabilis	0.008	0.016	0.125	0.125
Serratia marcescens	0.06	0.125	0.125	0.5
Pseudomonas aeruginosa	8	16	16	32
Enterobacter cloacae	16	32	16	32

Table IV

Time, hr	Aztreonam: *E. coli*	*Kleb. pneum.*	*Proteus mirabilis*	*Serratia marcescens*	*Pseudo. aeruginosa*	*Ent. cloacae*
Pre	<2	<2	<2	<2	<2	<2
0.5	144	45	57	57	6	6
2	72	18	28	32	2	2
6	16	10	7	57	<2	2
8	12	7	4	16	<2	<2
12	6	4	3	7	<2	<2
	Moxalactam					
Pre	<2	<2	<2	<2	<2	<2
0.5	45	60	32	72	2	9
2	28	23	20	45	<2	6
6	11	6	7	18	<2	2
8	8	3	5	8	<2	2
12	6	<2	2	2	<2	<2

Table V

Time, hr	Aztreonam: *E. coli*	*Kleb. pneum.*	*Proteus mirabilis*	*Serratia marcescens*	*Pseudo. aeruginosa*	*Ent. cloacae*
Pre	<2	<2	<2	<2	<2	<2
0-0.5	2048	1552	1552	676	184	64
0.5-2	2048	1625	2299	813	456	102
2-6	1149	813	575	406	128	40
6-8	456	256	362	161	64	7
8-12	322	203	144	72	28	3
	Moxalactam					
Pre	<2	<2	<2	<2	<2	<2
0-0.5	9195	2560	362	228	28	81
0.5-2	10321	6502	322	256	32	203
2-6	5161	2048	181	144	23	72
6-8	3251	1625	102	102	11	36
8-12	1149	512	64	64	4	18

The bactericidal activity of aztreonam in humans (Tables IV and V) supports, in patients with normal renal function, a 2-gram q12h intravenous dosage regimen for systemic infections due to *E. coli*, *K. pneumoniae*, *P. mirabilis*, and *S. marcescens* having MIC's of 0.06, 0.06, 0.06 and 0.06 ug/ml, respectively. Therapy of systemic infections due to the test strains of *P. aeruginosa* (MIC = 8 ug/ml) and *E. cloacae* (MIC = 16 ug/ml) would appear to require more frequent administration and perhaps higher doses of aztreonam, in patients with normal renal function. Uncomplicated urinary infections by the test organisms might be treatable with aztreonam using a 0.5-gram q12h intravenous dosage regimen, in patients with normal renal function. However, these suggestions should be considered tentative, pending results of ongoing therapeutic trials in patients.

Aztreonam and moxalactam, administered as single, 2000-mg intravenous doses to healthy male subjects, had similar safety, pharmacokinetic, and bactericidal activity profiles in the present study. However, comparison of safety and efficacy of these two compounds in infected patients awaits results of ongoing clinical trials.

} Concur. Formulation: aztreonam/L-arg (1.0/0.78)

DEPARTMENT: Department of Clinical Pharmacology
Division of Medical Affairs

SECTION:

DATE OF REPORT: March 31, 1982

NDA #50-860

NAME OF DRUG: Aztreonam (SQ 26,776)

TITLE

Report on Oral Bioavailability of Aztreonam in Healthy Male Subjects.
Study Protocol # 18557-7

AUTHORS:
Edward A. Swabb, Ph.D., M.D., May Frantz, Ph.D., and Michelle A. Stern, B.A.

INVESTIGATORS:
A. A. Sugerman, M.D., Medical Center at Princeton, Princeton, NJ; T.B. Platt, Ph.D., The Squibb Institute.

ABSTRACT

The objective of this study was to determine the absolute oral bioavailability of aztreonam and the relative bioavailability of two oral dosage forms in healthy male subjects. Doses of 500 mg of aztreonam were administered as an oral solution, as two 250-mg capsules, and as a 3-minute intravenous infusion to 15 healthy male subjects at 2-week intervals according to a 3-way crossover study design. Two additional subjects were enrolled, but did not complete the study. Samples of serum and urine were collected at frequent intervals during this study for measurement of aztreonam by microbiological assay. To assess the safety of aztreonam, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and at frequent intervals during the study.

Mean bioavailability parameters (± S.E.M.) obtained from serum level data are summarized on the following page:

PARAMETER	UNITS	ORAL SOLUTION	CAPSULE	INTRAVENOUS INFUSION
C_{max}	µg/ml	0.13 ± 0.02	0.14 ± 0.02	56.7 ± 1.5[a]
T_{max}	hours	2.08 ± 0.38	2.23 ± 0.44	0.08 ± 0.0[a]
$AUC_{0-16\ hr}$	µg x hr/ml	0.45 ± 0.08	0.46 ± 0.05	81.3 ± 2.5
Absolute Bioavailability	% of AUC for IV	0.55 ± 0.09	0.49 ± 0.06	100.0

[a] C_{max} occurred 5 minutes after completion of the infusion of the drug and T_{max} represented the initial blood sample drawn.

Mean values for maximal concentration in serum (C_{max}), the time to attain maximal concentrations in serum (T_{max}), and the areas under the serum concentration vs. time curves ($AUC_{0-16\ hr}$) for 500-mg doses of aztreonam administered as an oral solution or capsule were not significantly different, but were markedly different from values for the intravenous infusion. The absolute bioavailability of each oral formulation, defined as the ratio of AUC p.o./intravenous, was less than 1%.

The amount of the administered dose that was excreted in the urine as microbiologically active drug is summarized below:

COLLECTION TIME, HR	CUMULATIVE URINARY EXCRETION % OF DOSE		
	ORAL SOLUTION	CAPSULE	INTRAVENOUS INFUSION
0-[illegible]	0.265 ± 0.074	0.195 ± 0.023	55.6 ± 1.7
0-8	0.533 ± 0.061	0.407 ± 0.043	65.3 ± 1.6
0-48	0.675 ± 0.076	0.549 ± 0.064	67.8 ± 1.6

Mean values for cumulative urinary excretion of aztreonam for various collection times for the oral formulations were not significantly different, but were markedly less than values for intravenous infusion.

Thus, based on serum levels and urinary excretion data, the two oral formulations were found to be bioequivalent. The oral dosage forms had less than 1% absolute bioavailability, in comparison to the intravenous dose. If high serum and urine levels of aztreonam are desired, this compound should be administered parenterally.

Aztreonam was tolerated well by healthy male subjects; there were no apparent adverse reactions associated with the administration of aztreonam.

The lack of oral bioavailability of aztreonam suggests that the orally administered drug will reach the large intestine and possibly kill susceptible aerobic gram-negative bacteria. This suggests a potential application for aztreonam oral prophylaxis of serious aerobic gram-negative infections in immunocompromised hosts, such as patients undergoing cancer chemotherapy. Further study of the safety and efficacy of orally administered aztreonam will be required to confirm this possibility.

Note: For the intravenous infusion study dose similar, mean serum levels and [illegible]

TABLE 20

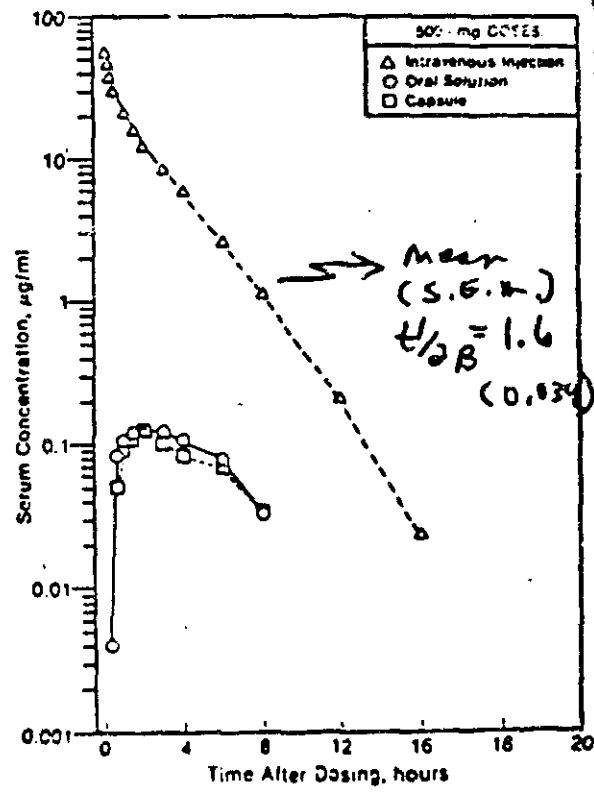
MEAN (± S.E.M.) CONCENTRATION (µG*/ML) OF AZTHREONAM IN THE SERUM OF NORMAL SUBJECTS AFTER ADMINISTRATION OF 500 MG AS AN ORAL SOLUTION, CAPSULE, AND 3-MINUTE INTRAVENOUS INFUSION

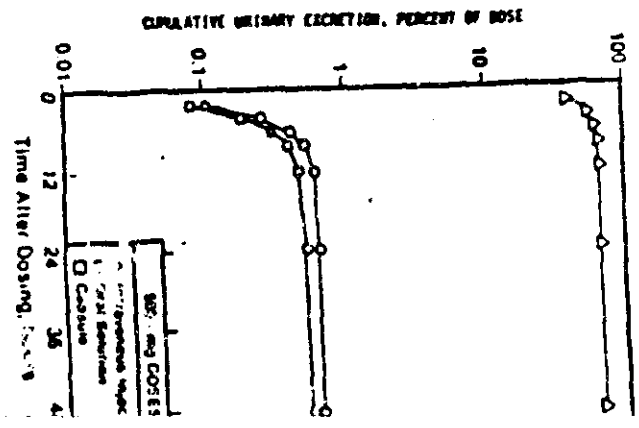
Time (hours)	Oral Solution	Capsule	Intravenous
0.08	NA[a]	NA	56.75 ± 1.46
0.17	0.000 ± 0.000	0.000 ± 0.000	46.48 ± 1.13
0.33	0.004 ± 0.004	0.000 ± 0.000	37.23 ± 0.87
0.50	NA	NA	30.34 ± 0.73
0.67	0.085 ± 0.015	0.053 ± 0.013	NA
1.0	0.108 ± 0.016	0.091 ± 0.017	20.76 ± 0.64
1.5	0.121 ± 0.018	0.110 ± 0.015	15.91 ± 0.57
2.0	0.125 ± 0.019	0.128 ± 0.010	12.22 ± 0.36
3.0	0.124 ± 0.018	0.103 ± 0.011	8.57 ± 0.34
4.0	0.108 ± 0.021	0.085 ± 0.011	5.92 ± 0.23
6.0	0.079 ± 0.016	0.068 ± 0.010	2.60 ± 0.12
8.0	0.033 ± 0.011	0.034 ± 0.012	1.11 ± 0.08
12.0	0.000 ± 0.000	0.000 ± 0.000	0.21 ± 0.02
16.0	0.000 ± 0.000	0.000 ± 0.000	0.02 ± 0.01

[a] NA = Not Applicable.

FIGURE 1

MEAN SERUM AZTHREONAM CONCENTRATION VS. TIME AFTER ADMINISTRATION OF 500 MG AS AN ORAL SOLUTION, CAPSULE, AND 3-MINUTE INTRAVENOUS INFUSION TO HEALTHY SUBJECTS





MEAN CUMULATIVE URINARY EXCRETION OF AZTHREONAM VS. TIME AFTER ADMINISTRATION OF 500 MG AS AN ORAL SOLUTION, CAPSULE, AND 3-MINUTE INTRAVENOUS INFUSION TO HEALTHY SUBJECTS

III. 3

Division of Medical Affairs	October 31, 1983
SECTION: Clinical Pharmacology	PROJECT CODE: MNB-860
	PRODUCT, SQ NO., OR PROJECT NAME: Aztreonam (SQ 26,776)

TITLE:
Report on the Pharmacokinetic Study of Aztreonam (SQ 26,776) in Healthy Elderly Volunteers

Study Protocol # 18554-68

AUTHOR(S):
William A. Creasey, D. Phil., Janice Lux, B S., M.P.H., and May Frantz, Ph.D.

INVESTIGATORS:
A. Arthur Sugerman, M.D., The Medical Center at Princeton, Princeton, NJ, and T.B. Platt, Ph. D., S. Wind, B.S., M.A. Leitz, B.A., and J. Kartan, B.S., The Squibb Institute for Medical Research, New Brunswick, NJ

ABSTRACT:

Aztreonam was administered by single 3-minute intravenous injection to 13 healthy male volunteers aged 65 to 75 years (mean = 68.1 yrs) at a dose of 1000 mg. One subject was dropped from the study because of failure to obtain blood samples; 12 subjects completed the study. Assays were performed for aztreonam and its metabolite SQ 26,992 in sera, protein-free filtrates of sera and urine samples using HPLC methodology. Selected serum samples (20 min and 8 hr) were assayed microbiologically for aztreonam; no discrepancies between the two methods of assay were noted. Serum samples were drawn at 5, 10, 20 and 30 minutes, and 1,2,3,4,6,8 and 12 hours after injection. Protein-free filtrates of sera were prepared from samples drawn at 10 minutes, 1 and 3 hours after injection. Urine was collected during the -8 to 0, 0 to 2, 2 to 4, 4 to 6, 6 to 8, 8 to 12, 12 to 24 and 24-48 hour periods relative to the end of injection. All subjects received physical examinations, 12-lead electrocardiograms, and a battery of tests of blood and urine chemistry, hematology and serum enzymes.

Elderly volunteers' Creatinine Clearance ranged from 70.9 to 163 ml/min (mean = 100 ml/min).

The tables below summarize the pharmacokinetic findings for aztreonam and the urinary excretion of SQ 26,992. Comparison of the data with that from a similar study (Protocol 18,554-1) carried out in an 18 to 35 year-old group of male volunteers showed similar maximum serum levels and volume of distribution. However, the distribution constants (K_{12} and K_{21}) were larger, and the serum clearance somewhat slower in the older subjects, although the net effect on elimination half-life and urinary excretion was probably not significant clinically.

PHARMACOKINETIC PARAMETERS FOR AZTREONAM (Normals)

Parameter	Units	Mean	S.E.M.	Range
C_{max}	µg/ml	120.1 (158 HPLC)	4.25	97.3 - 154.0
Serum-protein Binding at 10 min	percent	50.4	2.65	27.1 - 59.9
AUC 0-24 hr (trapezoidal)	µg.hr/ml	231.8 (331 Bioassay)	9.18	176.8 - 285.6
V_1	liters/kg	0.08 (0.07)	0.01	0.03 - 0.11
V_{ss}	liters/kg	0.15 (0.14)	0.01	0.12 - 0.22
V_{area}	liters/kg	0.16 (0.16)	0.01	0.13 - 0.23
$t_{\frac{1}{2}\alpha}$	hr	0.15 (0.22)	0.02	0.04 - 0.33
$t_{\frac{1}{2}\beta}$	hr	2.06 (1.85)	0.06	1.60 - 2.35
k_{10}	hr^{-1}	0.75 (0.81)	0.06	0.42 - 1.17
k_{12}	hr^{-1}	3.40 (4.38)	1.01	0.66 - 6.11
k_{21}	hr^{-1}	2.76 (1.63)	0.35	1.24 - 5.13
24 hour urinary excretion	%	63.1 (73.4)	1.84	51.9 - 73.5
Serum clearance	ml/min/kg	0.94 (0.99)	0.07	0.66 - 1.60

Urinary Excretion of SQ 26,992

Time (hr)	Mean (percent of dose)	S.E.M.
0 - 24	3.06	0.18
0 - 48	5.13	0.28

TABLE 3

MEAN SERUM CONCENTRATIONS (µg/ml) ± S.E.M. FOR AZTREONAM AT EACH SAMPLING TIME

TIME AFTER INJECTION hr	Aztreonam Concentration	
	HPLC	Microbiologic
0.08	120.1±4.3	ND
0.17	98.4±4.7	ND
0.33	80.6±3.0	83.2±1.1
0.5	69.9±2.0	ND
1.0	53.2±1.7	ND
2.0	36.1±1.2	ND
3.0	26.4±1.2	ND
4.0	18.9±1.0	ND
6.0	10.1±0.7	ND
8.0	5.3±0.5	5.8±0.5
12.0	1.6±0.3	ND

Concentration of Aztreonam and SQ 26,992 in Urine[a]

Time After Infusion, hr	Aztreonam	SQ 26,992
0-2	1388 ± 280	15.7 ± 5.2
2-4	935 ± 176	18.8 ± 5.8
4-6	718 ± 116	26.4 ± 5.4
6-8	363 ± 39	21.8 ± 2.5
8-12	175 ± 34	23.2 ± 2.8
12-24	40 ± 16	19.7 ± 1.9
24-48	[b]	11.6 ± 1.5
% of dose excreted in urine,		
0-24	63.1 ± 1.8	3.1 ± 0.2
0-48	[b]	5.1 ± 0.3

[a] Urinary concentration values were determined using an HPLC assay and are expressed as mean ± SEM in units of µg/ml; 12 subjects were studied.

[b] Only one subject excreted aztreonam in a detectible concentration

Pharmacokinetics of aztreonam in elderly patients with intra-abdominal infections (Addendum A to Protocol 18554-38)

Although this addendum study is still in progress, preliminary results have provided useful information on the age dependence of aztreonam disposition. These findings are described below.

The pharmacokinetics of aztreonam have been investigated in 3 female and 4 male Caucasian patients with ages less than 65 years (mean, 50 years; range, 18 to 61 years) and in 4 female and 2 male patients (5 Caucasian, 1 Negro) with ages greater than 65 years (mean, 82 years; range, 72 to 91 years). The former group had body weights ranging from 63.0 to 81.8 kg (mean, 72.0 kg), and the latter group had body weights ranging from 54.5 to 81.0 kg (mean, 71.7 kg). Two additional patients were enrolled in this addendum study but were excluded from analysis because of insufficient serum concentration data. All patients were being treated with aztreonam (2000 mg intravenously q6h to q24h) and clindamycin (600 mg intravenously q8h to q8h) for known or suspected intra-abdominal infections, according to Protocol 18554-38.

The following table indicates that elimination of aztreonam was significantly impaired in elderly patients (age greater than 65 years), as reflected by a 55% decrease in mean serum clearance of aztreonam, and a 240% increase in elimination half-life ($t_{1/2\beta}$). There was no important change in the apparent steady-state volume of distribution (V_{ss}).

n = 7 6

Parameter[a]	age <65 yr.	age >65 yr
serum clear., ml/min	105.7 ± 15.9	47.8 ± 13.0
V_{ss}, liters/kg	0.26 ± 0.04	0.27 ± 0.02
$t_{1/2\beta}$, hr	2.5 ± 0.4	8.5 ± 3.0
renal clear., ml/min	100 ± 8	40 ± 1[illegible]

[a]Values are mean ± SEM.

The apparent relationship between age and impaired elimination of aztreonam is explained in large part by the presence of significant renal insufficiency in the elderly patient population, which had a mean creatinine clearance of 40 ml/min, compared to 100 ml/min in patients less than 65 years of age. Figure 3 portrays these findings graphically, and provides the regression line obtained from a single intravenous-dose kinetic study in subjects with normal or impaired renal function (Protocol 18554-8) for comparison. Considering the variability in clearance values for aztreonam in patients enrolled in this addendum study, the age dependence of aztreonam pharmacokinetics can be explained by the age dependence of renal function. Dosage adjustment can be based directly upon creatinine clearance in adults of various ages.

Note: No raw data were provided initially for Protocol #18,554-38 Addendum A. In a meeting with the firm on 5/22/86 additional data were provided for this study (see III-4b and 4c).

Comments as Related to the firm's Summary

1) Study Protocol #18,554-6 (i.e., 3 minute IV infusion of 1000 mg drug; n = 13 healthy o's; aged 65 to 75 years; mean creatinine clearance 100 ml/min (range, 71-16) had PK mean values of:

Vss (L/kg) = 0.15 Range (0.12 - 0.
$t_{1/2\beta}$ (hr) = 2.06 (1.68 - 2.
Serum clearance (ml/min/kg) =

2) Study Protocol # 18,554-8 (i.e., 2 minute infusion of 1000 mg; n = 5 o's aged 34 to 54 years; mean creatinine clearance 45 ml/min (range 36-54 ml/min)) had PK values of:

Vss (L/kg) = 0.17 Range (0.14 - 0
$t_{1/2\beta}$ (hr) = 3.7 (2.7 - 4.5
Serum clearance (ml/min) = 48.7 (36.3 - 64.8)

3) The two studies described above tend to support the findings of study Protocol # 18,554-38 based upon creatinine and serum clearance.

DEPARTMENT:
Clinical Pharmacology

DATE OR PERIOD OF STUDY:
January 10, 1986

SECTION:
Division of Medical Affairs

PROJECT CODE:
MMB-860

PRODUCT, SQ NO., OR PROJECT NAME
AZTREONAM

TITLE: Report of a Study of Pharmacokinetics and Extravascular Penetration of Aztreonam in Patients With Abdominal Sepsis

AUTHOR(S): Lawrence I. Friedhoff, Ph.D., M.D., The Squibb Institute for Medical Research, Princeton, New Jersey 08540

INVESTIGATORS: J. Schentag, Pharm. D., P.B. Wels M.D. and J. Patel, M.D., Millard Fillmore Hospital, 3 Gates Circle, Buffalo, New York 14209

Study Protocol # 18554-38

ABSTRACT:

Addendum A continued

The attached documents represent the final report for Protocol 18554-38 Addendum A (entitled "Addendum A to Protocol 18554-38 for Dr. P. B. Wels"). The manuscript entitled "*Pharmacokinetics and Extravascular Penetration of Aztreonam in Patients With Abdominal Sepsis*" summarizes results for 20 patients (published in Rev. Infec. Dis. 7(Supp. 4):S716-S723, 1985). One additional patient listed in the manuscript (SG) had pneumonia and peritonitis and was enrolled in Protocol 18554-11 Addendum A. Exclusion of data from this patient has no significant effect on the mean age, weight, creatinine clearance, total body aztreonam clearance (TBC), V_{dss}, V_c, V_t or $t_{\frac{1}{2}}$. Mean values of these parameters calculated without the data of patient SG are listed below.

Age (yr)	Weight (kg)	TBC (ml/min)	V_{dss} (L/kg)	Vc (L)	V_t (L)	$t_{\frac{1}{2}}$ (hr)
62.9	69.3	81.1	0.28	9.4	9.1	4.68

Seven additional patients received aztreonam in the study, but these data were not included in the manuscript. Data for these 7 patients are listed in Table 1. The mean values were similar to those of the first 20 patients. Aztreonam levels in body fluids are listed in Table 2.

The safety data for these patients have been reported with results of Phase II-III studies in the New Drug Application filed with the FDA on December 28, 1983 and The Aztreonam Safety Update of December 1985 issued by S.S. Tadros, T.J. Newman, G.R. Dreslinski, M.D. Barnhart, C. Nagan and J.R. Odell.

New data provided by the firm on 5/22/86 at an FDA meeting with HFN-515.

Protocol 18554-38
Addendum A

TABLE 2

Pt. No.	Treatment Regimen Prior to Sampling of Abdominal Fluid	Abdominal Fluid Sampled[a]	Time From Last Dose, hr	Aztreonam Conc. (μg/ml or μg/gm)
206	2gm IV q6h x 8 days	peritoneal	2.50	34.5
207	2gm IV q6h x 1 day	subphrenic abscess	3.75	<0.3
		drain	3.75	12.5
		drain	1.3	0.0
210	2gm IV q12h x 3 days	drain	3.5-6.5	29.4
		drain	0-1	43.7
		drain	1-3	45.1
		drain	3-5	46.3
		drain	5-6	86.3
211	1gm IV x 1 dose	pus	1.5	10.1
215	2gm IV q24h x 2 days	bile	6.33-8.5	33.5
		drain	6.25-8.5	7.8
219	1gm IV x 1 dose	pus	3.75	12.0
	2gm IV q8h x 6 days, then 1gm IV	drain	0-3	[illegible]
		drain	3-6	99
		drain	6-7.5	83
222	2gm IV x 1 dose	pus	0.0	76.7
	2gm IV q12h x 3 days	drain	0-6.2	114.5
224	1gm IV x 1 dose	bowel	2.7	2.7
	2gm IV q8h x 3 days	drain	0.5	19.8
225	2gm IV q8h x 6 days	drain	0	7.6
	2gm IV q8h x 3 days	subphrenic abscess	6.25	6.8
	2gm IV q8h x 6 days	drain	3-6	2.0
230	2gm IV q24h x 9 days	drain	23-26	11
		drain	0-1.75	48.7
		drain	1.75-3.75	66.6
		drain	3.75-5.75	49.1
		drain	5.75-8.75	61.5
		drain	8.75-10.75	33.6
		drain	10.75-20.75	39.1

[a]Peritoneal = Non-infected fluid aspirated from the peritoneal space during surgery; Pus = Infected fluid aspirated from the peritoneal space during surgery; Subphrenic abscess = Loculated infected fluid aspirated from the subphrenic space during surgery; Drain = Fluid obtained after surgery from a drain left in the peritoneal space; Bile = Bile obtained from a T-tube after surgery.
When samples were collected over an interval rather than at a single time, the entire interval is listed as the sampling time.

TABLE 1

SUMMARY OF AZTREONAM STEADY-STATE PHARMACOKINETIC PARAMETERS AS DETERMINED BY TWO COMPARTMENT ANALYSIS

PATIENT	AGE/SEX	WEIGHT (kg)	CREATININE CLEARANCE (ml/min)	A (μg/ml)	α (hr^{-1})	B (μg/ml)	β (hr^{-1})	TBC (ml/min)	V_{dss} (L/kg)	V_c (L)	V_t (L)	$t_{\frac{1}{2}}$ (hr)	K_{el} (hr^{-1})	K_{21} (hr^{-1})	K_{12} (hr^{-1})
[redacted]	32/M	107													
	72/M	51													
	64/M	85													
	57/M	79													
	73/F	84													
	91/M	80													
	MD/MD	MD													
MEAN[a]	60	88	62.3	92.28	2.69	45.94	0.29	333.6	0.37	15.0	16.8	4.72	0.45	1.6.	1.34
SEM[a]	117	11	31.2	29.59	1.27	13.36	0.09	61.8	0.11	3.1	8.0	2.59	0.22	0.59	0.89

[a]excluding patient AS and RS.

[b]CL=1.45[creatinine clearance]+22.6

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Pharmacokinetics and Extravascular Penetration of Aztreonam in Patients with Abdominal Sepsis

Nancy E. Wiedsma, Ian L. Smith, Gary W. Simons, Douglas J. Swanson, Alessandro Vigano, Philip B. Weis, and Jerome J. Schentag

From the Department of Pharmaceutics and Division of Clinical Pharmacy Sciences, School of Pharmacy, State University of New York at Buffalo, and the Departments of Surgery and the Clinical Pharmacokinetics Laboratory, Millard Fillmore Hospital, Buffalo, New York

Patients with abdominal sepsis were enrolled in a clinical trial of aztreonam vs. tobramycin. All were given clindamycin concommitantly. The pharmacokinetics of aztreonam in 21 patients randomly assigned to receive treatment with aztreonam are reported. The mean age of these patients was 68 years; most had underlying disorders such as malnutrition and cardiac or pulmonary disease. Creatinine clearance (Cl_{cr}) ranged from 11.2 to 133.1 ml/min. The usual dose of aztreonam was 2.0 g every 8–12 hr. A single pharmacokinetic study was performed over one dosing interval after steady-state conditions were achieved. In approximately one-half of the patients, peritoneal fluid was collected during the interval between doses. Penetration of aztreonam as expressed as the ratio of concentration in the peritoneal fluid to that in serum, was higher for aztreonam (0.95:1) than for tobramycin (0.44:1). The ratio of the concentration in peritoneal fluid to the minimum inhibitory concentration (MIC) of the infecting bacteria was also higher for aztreonam. Serum pharmacokinetic data were analyzed by both two-compartment and moment analysis. For both the steady-state volume of distribution (Vd_{ss}) and total body clearance (TBC), the values determined by both methods were highly correlated ($r = .96$, .99, respectively). Average values for Vd_{ss} and TBC were 0.28 liters/kg and 80 ml/min. TBC for aztreonam correlated strongly with Cl_{cr} and was described by the regression equation $TBC = 1.1\ (Cl_{cr}) + 1.6$, $r = .87$, $P < .01$.

assessed. The concentrations achieved were compared with the MICs of aztreonam for the bacteria isolated from these patients.

Patients and Methods

Patients. All patients were enrolled in a randomized comparison of tobramycin (Dista Products, Indianapolis) and aztreonam (E. R. Squibb & Sons, Princeton, N.J.)—each administered with clindamycin (Upjohn, Kalamazoo, Mich)—in the treatment of abdominal sepsis. All patients had generalized peritonitis at the time of entry into the study. Men and women 18 years of age or older were enrolled if they had presumptive evidence of intraabdominal infection, such as peritonitis with rebound tenderness, presence of free air on radiographic examination, fever, and leukocytosis and required immediate surgery. Exclusion criteria included pregnancy, granulocytopenia, a history of severe allergic reaction to penicillin, or an infection caused by bacteria resistant to either aztreonam or tobramycin in vitro. Written informed consent was obtained from all patients for both the use of aztreonam for the treatment of the abdominal infection and for the pharmacokinetic studies.

Most patients underwent surgery immediately after receiving the first dose of either aztreonam or tobramycin. Thereafter, patients receiving aztreonam were given 2.0 g every 8–12 hrs and those receiving tobramycin received a dose that would result in measured peaks of 4.0–10.0 μg of tobramycin/ml and in troughs of <2.0 μg/ml. On one occasion, at least three days after the initiation of aztreonam treatment, a pharmacokinetic study was performed over the dosing interval. The dose of aztreonam was reconstituted with sterile water for injection and was then diluted with 10–20 ml of 5% dextrose in water. It was administered iv over an interval of 3 min. Venous blood samples were drawn before administration of the antibiotic and at 10, 20, 30, and 45 min, and 1, 1.5, 2, 4, 6, and 8 hrs after the 3-min infusion was complete. Blood samples were allowed to clot and the serum was removed and buffered to pH 6.0 with phosphate buffer. Samples were frozen at −20°C pending assay.

In approximately one-half of the patients, peritoneal fluid was collected during the interval between administrations of antibiotic, and the concentrations of aztreonam or tobramycin in this fluid were measured with use of serum standard curves. In several patients, cumulative samples of abdominal fluid drainage were collected over one dosing interval, thereby allowing the construction of a graph of concentration of antibiotic vs. time for peritoneal fluid. Penetration of antibiotic into abdominal fluid was assessed by determining the ratio of the concentration in abdominal fluid to that in serum and by comparing the concentration in abdominal fluid to the MIC for the infecting bacteria.

Aztreonam was analyzed by reversed-phase high-performance liquid chromatography (HPLC) in which a mobile phase of acetonitrile, tetrabutylammonium hydrogen sulfate, and water was employed. Column effluent was monitored at 254 nm, and peaks were analyzed with reference to the internal standard cefoxitin by use of area integration. The procedure was shown to be highly specific for aztreonam and sensitive to a concentration of 2.0 μg/ml. The HPLC assay also quantitates the principal aztreonam degradation product, SQ 26,992. Tobramycin was assayed with use of an enzyme immunoassay (EMIT; Syva, Palo Alto, Calif).

Pharmacokinetic analysis. Data on serum concentration vs. time were fitted to a two-compartment, open mamillary model with use of the damping Gauss-Newton nonlinear least-squares algorithm [10]. This program was adapted to a Tektronix 4052 microcomputer (Beaverton, Ore.). The derived parameters were mathematically corrected to values

Table 1. Clinical characteristics of 21 patients receiving aztreonam for treatment of abdominal sepsis.

Characteristic	Value
Age (years)	
Mean	68.1
Range	18–91
Sex (M/F)	9/12
Weight (kg)	
Mean	69.3
Range	44–111
Creatinine clearance (ml/min)	
Mean	69.3
Range	11–133
Infection site (no. of patients)	
Upper gastrointestinal tract	5
Hepatobiliary	2
Jejunum/ileum	5
Colon	3
Appendix	5
Other	1
Combinations	4

Results

Demographic data for the 21 study patients are provided in table 1. Many patients had multiple underlying diseases, the most common being malnutrition and cardiac disease. Chronic obstructive pulmonary disease was often frequent; hepatic disease and neoplasia were encountered less frequently. The initial gram-negative isolates for the 10 patients for whom concentrations of aztreonam in the abdominal fluid were measured for nine patients in this study, and the MICs for the infecting organisms were determined (table 2).

Figure 1 illustrates the average time course of serum concentrations of aztreonam for patients with

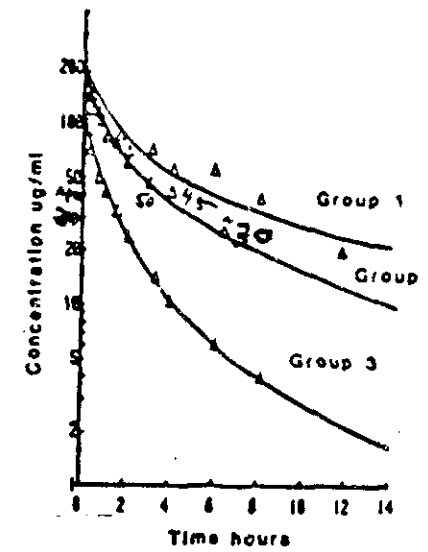

Figure 1. Semilogarithmic plots of serum concentration of aztreonam vs. time in patients with different levels of renal function. For patients in group 1, creatinine clearance (Cl_{cr}) was 0–40 ml/min; for those in group 2, 40–80 ml/min; and for those in group 3, >80 ml/min (normal). Values for representative patients were used in constructing these plots.

normal renal function (Cl_{cr} >80 ml/min) and for two groups of patients with decreased renal function. The biexponential characteristic of these data requires the use of a two-compartment model to describe the steady-state disposition of aztreonam. A summary of the pharmacokinetic parameters as determined by two-compartment analysis is provided in table 3.

Figure 4 illustrates the relation between TBC and Cl_{cr} for the two-compartment model. Cl_{cr} therefore accounted for 75% ($r^2 \times 100$) of the observed variability in the TBC of aztreonam.

Two-compartment analysis yielded a t½ value of 4.6 hr, whereas moment analysis yielded a serum t½ value of 4.5 hr. Serum t½ of aztreonam was inversely related to Cl_{cr}. When the average t½ was calculated for patients grouped according to renal function, the average serum t½ for those whose Cl_{cr} was >80 ml/min was 2.6 hr. For patients whose Cl_{cr} was 40–80 ml/min, the average serum t½ was 4.1 hr, and for those whose Cl_{cr} was <40 ml/min, the average t½ was 8.7 hr.

Figure 5 illustrates the concentrations of aztreonam in both the serum and peritoneal fluid of a representative patient. The peak concentration in peritoneal fluid occurred later than did the peak in serum; these peaks declined in parallel within 2–3 hr of dosing. The average ratio of the concentration of aztreonam in peritoneal fluid to that in serum was 0.95:1.

The ratio of the concentration of aztreonam or tobramycin in the abdominal fluid and the respective MICs for the gram-negative isolates are shown in figure 7. The average ratio for aztreonam was 12.9 and that for tobramycin was 2.2.

Table 2. Initial gram-negative isolates for 21 patients receiving [illegible] for treatment of abdominal sepsis and the MICs for the isolates.

Organism	Tobramycin No. of isolations	Tobramycin MIC	Aztreonam No. of isolations	Aztreonam MIC
Escherichia coli	5	0.6 ± 0.2*	4	1.3 ± 1.0*
Pseudomonas aeruginosa	3	0.5 ± 0.0*	3	5.2
Enterobacter species	2	2.0	1	0.2
Klebsiella species	1	1.0	3	1.9 ± 1.2*
Citrobacter	0	...	1	12.5
Proteus species	3	0.8 ± 0.3*	3	6.2
Serratia species	1	9.5	0	...

* Expressed as mean MIC ± SD.

Table 3. Summary of steady-state pharmacokinetic parameters for aztreonam in 21 patients as determined by two-compartment analysis.

Patient no.	Age/sex	Wt (kg)	Cl_{cr} (ml/min)	A (μg/ml)	α (hr⁻¹)	B (μg/ml)	β (hr⁻¹)	TBC (ml/min)	Vd_{ss} (liters/kg)	V_c (liters)	V_γ (liters)	t½ (hr)	k_{el} (hr⁻¹)	k_{12} (hr⁻¹)	k_{21} (hr⁻¹)
1															
2															
3															
4															
5															
6															
7															
8															
9															
10															
11															
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19															
20															
21															
Mean	68.1	69.1	69.3	200.73	4.29	80.77	0.23	79.5	0.28	9.8	9.2	4.61	0.62	1.22	2.60
SD	19.8	14.4	33.4	236.49	1.05	31.49	0.11	37.6	0.08	4.0	4.8	4.32	0.41	0.76	3.12

NOTE. Results are rank ordered by creatinine clearance (Cl_{cr}) values. A = y intercept of α phase; B = y intercept of β phase; α = rate constant for distribution phase; β = rate constant for elimination phase; TBC = total body clearance; Vd_{ss} = volume of distribution at steady state; V_c = volume of distribution of central compartment; V_γ = volume of distribution of tissue compartment; t½ = serum half-life; k_{el} = first-order elimination rate constant; k_{12} = first-order intercompartmental transfer rate constant from the central compartment to the peripheral compartment; k_{21} = first-order intercompartmental transfer rate constant from the peripheral compartment to the central compartment.

Discussion

The aztreonam serum concentration vs. time profiles for the study patients with normal renal function were similar to those found in a study of healthy volunteers [6]. The average elimination-phase t½ (4.5 hr) in our patients was, however, longer than that measured previously in volunteers (1.7 hr) [6]. This difference in serum t½ can be attributed primarily to the older age and reduced renal function of the majority of our patients (table 1). If our study patients whose Cl_{cr} was >80 ml/min are considered separately, the average t½ in this group (2.6 hr) is similar to that for the healthy volunteers. The Vd_{ss} for aztreonam (0.28 liters/kg) was larger than the Vd_{ss} (0.18 liter/kg) determined for healthy male volunteers. In addition to the physiologic differences between patients and healthy volunteers, variability in mathematical calculation of pharmacokinetic parameters and differences in protein binding for these two groups [7] may account for the differences in Vd_{ss} values.

This analysis indicated that Cl_{cr} was the most important determinant of the TBC of aztreonam. The dependency of aztreonam clearance on Cl_{cr} was expected, since the recovery of 70% of a single iv dose in the urine within 12 hr has been reported [5]. Both glomerular filtration and renal tubular secretion appear to be involved in the excretion of aztreonam [16].

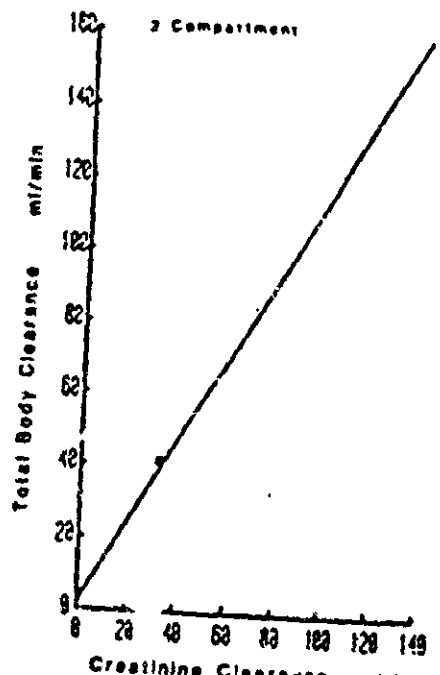


Figure 4. Total body clearance of aztreonam (TBC) vs. creatinine clearance (Cl_{cr}) for 21 patients. The relationship for two-compartment analysis is described by the regression equation TBC = 1.6 + 1.1 (Cl_{cr}), r = .87; P < .01. For moment analysis, the equivalent relationship is described by the equation TBC = 3.2 + 1.2 (Cl_{cr}), r = .85; P < .01.

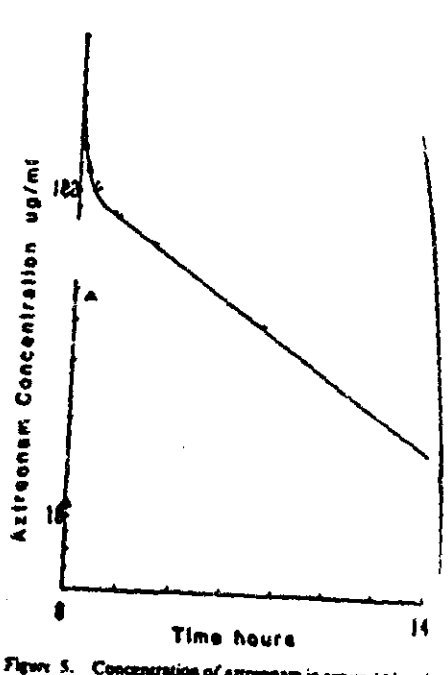


Figure 5. Concentration of aztreonam in serum (Δ) and peritoneal fluid (▲) vs. time in a representative patient.

5. Swabb EA, Leiz MA, Pilkiewicz FG, Sugerman AA. Pharmacokinetics of the monobactam SQ 26,776 after single intravenous doses in healthy subjects. J Antimicrob Chemother 1981;8(Suppl E):131-40

6. Swabb EA, Sugerman AA, McKinstry DN. Multiple-dose pharmacokinetics of the monobactam aztreonam (SQ 26,776) in healthy subjects. Antimicrob Agents Chemother 1983;23:125-32

7. Reitberg DP, Cumbo TJ, Smith IL, Scheetz JJ. Effect of protein binding on ±/±monosine steady-state kinetics in critical patients. Clin Pharmacol Ther 1984;35:64-71

16. Swabb EA, Sugerman AA, Frantz M, Plitt TB, Stern M. Renal handling of the monobactam aztreonam in healthy subjects. Clin Pharmacol Ther 1983;33:609-14

Protocol # 18,554-

Protocols # 18,554-, 18,554-

Protocol # 18,554

Additional Comments

This study (# 18,554-38) further provides data demonstrating the effect of decreased renal function (as measured by Cl_{cr}) on observed aztreonam serum levels. Comparison of the observed "mean" steady-state drug levels for aztreonam given as 2g every 8-12 hours intravenously (Figure 1, III.4C) with the corrected (predicted) drug levels discussed in Section IV of this review (4/30/86 memo to Dr. Min), suggest that the predicted and observed levels for the two renally impaired groups to be reasonably close. Study # 18,554-38 also demonstrates that for patients with Cl_{cr} within the range of 30-80 ml/min/1.73 m² (actual value 43-79 ml/min) the mean steady-state drug levels are much closer than

Department of Clinical Pharmacology | August 10, 1983

SECTION: Division of Medical Affairs | PROJECT CODE: MRB-660 | PRODUCT, IND NO., OR PROJECT CODE: SQ 26,776 (Aztreonam)

TITLE:

Report on Intravenous Safety and Pharmacokinetic Study of Aztreonam (SQ 26,776) in Patients with Stable Chronic Renal Failure

Study Protocol # 18,554-24

Formulation: aztreonam/L-arginine (1.0/0.78)

AUTHORS: Edward A. Swabb, Ph.D., M.D., and Cecelia Verrucci, B.A.

INVESTIGATORS: Jean-Paul Villastre, M.D., Department of Nephrology, University of Rouen, School of Medicine, Rouen, France; Mr. C. Baudouin, Laboratories Squibb, Epernon, France.

ABSTRACT

Aztreonam was administered by single, 2-minute, 1000-mg intravenous infusion to 25 male or female volunteers with normal renal function (n=5) or with various degrees of renal insufficiency (n=20). To assess the safety of aztreonam, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and after drug administration.

The pharmacokinetic profile of aztreonam was assessed by measuring aztreonam and SQ 26,992 concentrations in multiple serum and urine samples collected from each subject. Microbiological and high-pressure liquid chromatography (HPLC) methods were used by the clinical investigator and Squibb analysts, respectively, to assay each sample. HPLC results were consistent with previously reported aztreonam studies and were, therefore, chosen as the basis for the pharmacokinetic analysis. Study volunteers were grouped into five categories of renal function for purposes of data analysis, as shown in Table I.

Renal insufficiency markedly delayed the elimination of aztreonam, as indicated by the pharmacokinetic data in Table I. Single 1000-mg intravenous doses produced serum and urine concentrations that would be bactericidal to commonly encountered members of Enterobacteriaceae and Pseudomonas aeruginosa. Even patients with non-zero clearances below 10 ml/min had potentially therapeutic levels of aztreonam in their urine.

This was an open study in which each patient in Groups I-I[V] received a single 1000-mg intravenous dose of aztreonam. [T]he patient in Group V received a single 1000-mg intravenous d[ose] during an interdialytic period (at least 48 hours prior to dialysis treatment) and a second 1000-mg dose immediately prior to hemodialysis, with 3 to 15½ weeks between doses.

Table I

Renal Function and Serum Pharmacokinetics of Aztreonam[a]

Parameter	Degree of Renal Insufficiency				
	Normal	Mild	Moderate	Severe: Not Req. HD	Severe: Requiring HD
Number of Patients	5	5	6	4	5 → Received dose at least 48 hours prior to dialysis treatment.
Range of Creatinine Clearance Values (ml/min)	91-137	35-61	13-24	6-9	anuric
Grouping	>50	30-80	10-29	2-9	<2
Serum Conc: (µg/ml)					
Time After = 0.17 hour	91 ± 3	103 ± 8	103 ± 4	93 ± 12	90 ± 11
Dosing 1	44 ± 2	72 ± 3	69 ± 3	69 ± 8	70 ± 5
6	7.3 ± 0.4	22 ± 4	33 ± 3	44 ± 5	39 ± 3
8	1.3 ± 1.3	14 ± 3	26 ± 3	37 ± 6	33 ± 4
12	0	7.2 ± 1.8	16 ± 2	27 ± 3	23 ± 4
24	--	0.2 ± 0.2	3.1 ± 1.1	8.5 ± 1.4	8.8 ± 2.3
48	--	0	0	0.6 ± 0.3	1.5 ± 0.6
AUC, µg hr/ml 0-48	182 ± 11	451 ± 69	615 ± 55	885 ± 92	833 ± 91
Urine Conc: (µg/ml)					
Collection = 0-2 hour	1265 ± 520	677 ± 200	392 ± 73	17[illegible] ± 36	--
Interval 8-12	57 ± 16	103 ± 24	120 ± 32	91 ± 9	--
12-24	1.0 ± 0.6	39 ± 9	42 ± 12	41 ± 7	--
24-48	0	0.5 ± 0.5	17 ± 10	11 ± 4	--
Cum. Excr. % of dose	60 ± 2	38 ± 2	24 ± 4	13 ± 1	--

} Also see Table 2

[a]All values, except creatinine clearance, are mean ± SEM. HD = hemodialysis.

The serum pharmacokinetics of aztreonam could be described by an open, linear, two-compartment kinetic model. Pharmacokinetic parameters for the five groups of patients are given in the next table. Renal insufficiency affected primarily the rate and extent of elimination of aztreonam, without having any meaningful effect on the rate and extent of aztreonam distribution. Small amounts of the open beta-lactam ring hydrolysis product of aztreonam, SQ 26,992, were found by HPLC assay in the urine of all patients.

[An ABC 11 u capillary tube coil hemodialysis machine with a 0.98 sq m surface area, blood flow rate 260-300 ml/min, and dialysate flow rate approximately 500 ml/min was used for 3 patients. During 4 hours of hemodialysis, the half-life of aztreonam in serum was 2.52 ± 0.31 hours, and 46 ± 4% of a prior 1000-mg dose was eliminated.]*

Aztreonam was tolerated well by all study subjects, and no drug-related adverse reactions were noted.

The linear regression equation for serum clearance of aztreonam, $Cl_{Az,s}$ (ml/min), vs. creatinine clearance, Cl_{Cr} (ml/min), was:

$$Cl_{Az,s} = 0.612\ Cl_{Cr} + 19.0,\ r = 0.912.$$

} Equation for Study Protocol # 18,554-8 was $Cl_{Az,s} = 0.609\ Cl_{Cr} + 25.2$, $r = 0.968$

Using this relationship, guidelines for modification of aztreonam dosage regimens were derived according to two methods: 1) reduction in dose while continuing to use a fixed dosage interval, and 2) increase in the dosage interval, while using a fixed dose. Dosage recommendations based on data obtained in this study were identical to those cited previously in the discussion of results of Protocol 18554-8.

Pharmacokinetic Analysis of Serum Aztreonam Data

Parameter	Degree of Renal Insufficiency				
	Normal	Mild	Moderate	Severe: Not Req. HD	Severe: Requiring HD
Distribution (Clcr)	91-137	35-61	13-24	4-9	anuric
• Extent					
V_1, liters/kg	0.12 ± 0.01	0.12 ± 0.02	0.12 ± 0.01	0.13 ± 0.01	0.20 ± 0.02
V_{ss}, liters/kg	0.20 ± 0.01	0.17 ± 0.01	0.19 ± 0.01	0.24 ± 0.02	0.27 ± 0.03
V_{area}, liters/kg	0.22 ± 0.01	0.19 ± 0.02	0.19 ± 0.01	0.24 ± 0.02	0.27 ± 0.02
• Rate					
$t_{\frac{1}{2}\alpha}$, hr^{-1}	0.18 ± 0.02	0.42 ± 0.20	0.26 ± 0.04	0.14 ± 0.04	0.68 ± 0.20
k_{12}, hr^{-1}	1.58 ± 0.37	2.19 ± 1.71	1.12 ± 0.23	2.90 ± 0.39	0.43 ± 0.17
k_{21}, hr^{-1}	2.32 ± 0.31	1.91 ± 0.50	1.81 ± 0.20	3.45 ± 0.78	1.13 ± 0.36
Elimination					
• Extent					
48-hr urinary excr., % of dose[a]	60 ± 2	38 ± 2	24 ± 4	13 ± 1	0
serum clear., ml/min	91.4 ± 6.2	43.1 ± 5.4	29.8 ± 2.9	20.4 ± 2.1	22.0 ± 3.1
renal clear., ml/min	54.5 ± 3.6	16.2 ± 2.3	6.8 ± 1.2	2.7 ± 0.4	0
non-renal clear., ml/min	36.9 ± 3.4	26.8 ± 3.4	23.0 ± 3.1	17.7 ± 1.8	22.0 ± 3.1
• Rate					
$t_{\frac{1}{2}\beta}$, hr^{-1}	1.82 ± 0.05	3.43 ± 0.35	5.31 ± 0.41	7.88 ± 0.60	8.40 ± 1.43
k_{10}, hr^{-1}	0.69 ± 0.03	0.39 ± 0.09	0.22 ± 0.01	0.16 ± 0.01	0.13 ± 0.03

[a] In comparison, the HPLC assay gave values of renal excretion of SQ 26,992 of 3.7 ± 2.5, 6.3 ± 1.6, 4.2 ± 0.4, and 3.4 ± 1.4 % of dose, respectively.

The results for this study support the findings in Study Protocol # 18,554-8.

See section IV of this review where this study is discussed further.

TABLE II

INDIVIDUAL CONCENTRATIONS OF AZTREONAM (ug/ml, BY HPLC ASSAY) IN ARTERIAL AND VENOUS SERUM AND DIALYSATE DURING HEMODIALYSIS[a]

SPECIMEN	TIME, HR	GROUP I - PATIENT NUMBER 11	12	13	14	15	MEAN ± SD
arterial serum	0.5						70.9 ± 4.3
	1						53.3 ± 3.3
	2						46.2 ± 2.3
	3						—
	4						24.7 ± 3.2
	6						11.4 ± 3.0
venous serum	0.5						6[illegible] ± 3.5
	1						52.4 ± 3.7
	2						37.1 ± 2.8
	3						—
	4						22.4 ± 2.1
	6						14.7 ± 4.6
dialysate	0.5						5.4 ± 0.3
	1						4.2 ± 0.3
	2						3.0 ± 0.2
	3						—
	4						1.8 ± 0.2
	6						0.9 ± 0.1

[a] BRL 16,992 was not measurable in all specimens.

Table III Guidelines for Dosage Modification for Patients with Renal Insufficiency

Method of Modification of Dosage Regimen	Degree of Renal Insufficiency[a]			
	Normal	Mild	Moderate	Severe[c]
Variable Dose, Constant Dosage Interval				
Dose, fraction of normal[b]	1	1/2	1/3	1/4
Dosage Interval, fraction of normal	1	1	1	1
Variable Dosage Interval, Constant Dose				
Dose, fraction of normal	1	1	1	1
Dosage Interval, multiple of normal	1	2	3	4

[a] Renal function was defined by creatinine clearance as follows: normal, >80 ml/min; mild, 30-80 ml/min; moderate, 10-29 ml/min; severe, <10 ml/min.

[b] Initial dose should be a loading dose equal to the normal dose.

[c] Patients requiring hemodialysis should receive half their usual (adjusted) dose after dialysis, to compensate for drug cleared by the procedure.

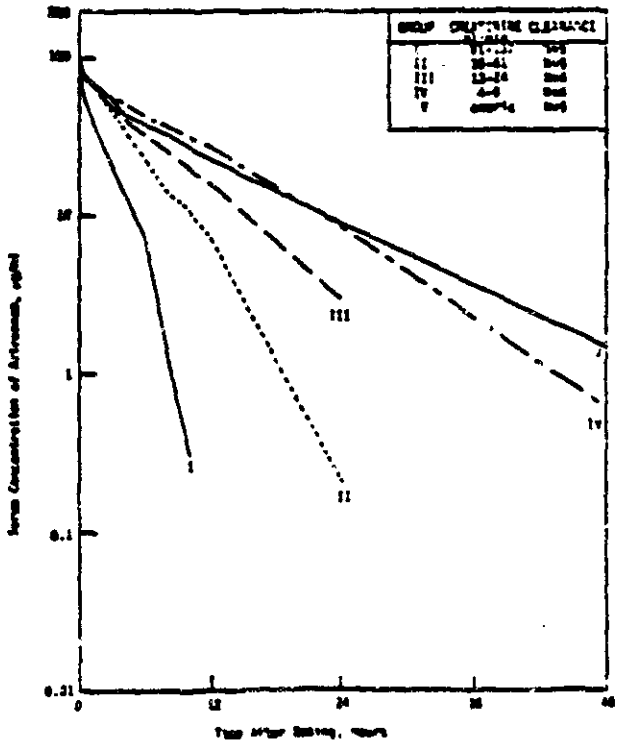
FIGURE 1

Effect of Renal Insufficiency on Serum Pharmacokinetics (HPLC assay) of 1-Minute Intravenous Infusions of 1000 mg Aztreonam

TABLE I

SERUM CONCENTRATIONS (MEAN ± STD, uGM/ML) OF AZTREONAM AFTER A SINGLE INTRAVENOUS DOSE OF 1000 MG USING HPLC ASSAY

TIME AFTER INFUSION HR	GROUP I (n=5)	GROUP II (n=5)	GROUP III (n=4)	GROUP IV (n=4)	GROUP V (n=5)
0.17	91.8 ± 3.2	103.0 ± 7.7	103.2 ± 4.4	93.8 ± 11.7	89.8 ± 10.4
0.33	69.4 ± 2.8	91.1 ± 3.8	87.5 ± 3.4	82.8 ± 11.1	83.1 ± 6.9
0.50	61.9 ± 3.2	86.8 ± 1.8	79.2 ± 2.9	76.8 ± 9.9	72.6 ± 9.0
0.75	52.4 ± 3.2	73.8 ± 3.7	75.1 ± 2.9	70.9 ± 7.9	77.7 ± 5.2
1	44.4 ± 2.6	72.3 ± 2.8	69.0 ± 3.4	66.6 ± 8.1	68.7 ± 5.8
2	30.4 ± 2.2	54.3 ± 4.3	56.6 ± 2.3	63.8 ± 8.2	62.7 ± 4.2
3	19.6 ± 1.6	42.9 ± 4.5	47.8 ± 2.6	56.7 ± 7.8	59.1 ± 3.7
4	14.6 ± 1.3	35.6 ± 4.2	43.1 ± 2.6	53.8 ± 6.7	62.9 ± 5.3
6	7.2 ± 0.4	21.7 ± 3.6	32.8 ± 2.[illegible]	43.7 ± 5.3	38.3 ± 3.3
8	1.3 ± 1.3	14.3 ± 3.8	23.9 ± 2.9	36.6 ± 5.9	33.1 ± 3.9
10	0.3 ± 0.3	10.7 ± 2.3	19.7 ± 2.1	28.2 ± 4.6	27.9 ± 3.5
12	0	7.2 ± 1.8	16.1 ± 1.9	27.2 ± 3.8	23.9 ± 4.1
24	—	0.2 ± 0.2	2.1 ± 1.1	9.5 ± 1.4	8.8 ± 2.3
48	—	0	0	0.6 ± 0.3	1.5 ± 0.6



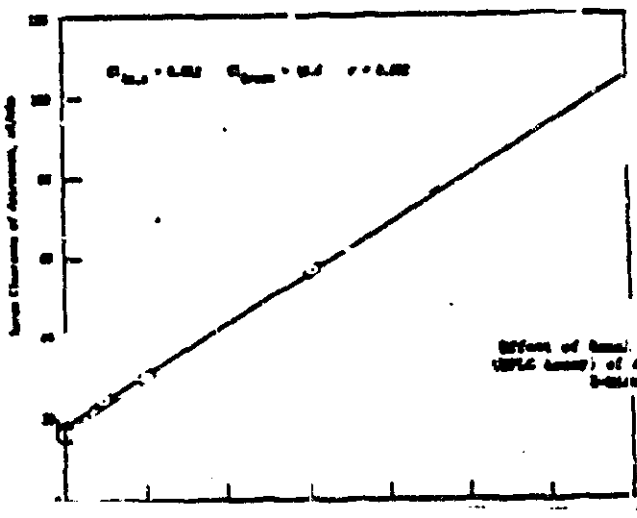


FIGURE 2

Effect of Renal Insufficiency on Serum Clearance (HPLC Assay) of Aztreonam Administered as a 1000-mg 1-Minute Intravenous Infusion

Pharmacokinetics and safety of aztreonam in patients on chronic hemodialysis or chronic ambulatory peritoneal dialysis (Protocol 18554-25)

Aztreonam was administered to 6 chronic hemodialysis (HD) patients with severe renal insufficiency as two 2-minute, 1000-mg intravenous infusions separated by a 1-week washout period. Each patient received one dose during the interdialytic period at least 48 hours prior to the scheduled hemodialysis and another dose 1 hour prior to hemodialysis. Aztreonam was also given to 6 patients with severe renal insufficiency and undergoing continuous ambulatory peritoneal dialysis (CAPD). These patients received a 1000-mg dose of aztreonam by a 2-minute intravenous infusion and another dose (separated by at least 1 week) by mixing 1000 mg of the antibiotic with a fresh 2-liter volume of peritoneal dialysis fluid just prior to fluid exchange. An additional CAPD patient left the study after receiving only one dose, and was replaced by another CAPD patient to bring the total to 6 completing the study.

To assess the safety of aztreonam, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and after drug administration.

The 9 male and 3 female subjects (8 Caucasian, 4 Negro) completing this study ranged in age from 20 to 64 years (mean, 42 years) and in body weight from 42.1 to 91.0 kg (mean, 65.4 kg).

Aztreonam was tolerated well by all patients, and no adverse reactions were noted.

The pharmacokinetic profile of aztreonam was assessed by measuring aztreonam concentrations in multiple serum, urine, and dialysate samples collected from each patient. A microbiological method was used to assay each sample. Serum and dialysate concentrations are summarized for hemodialysis patients and CAPD patients in the following two tables.

Hemodialysis Patients[a]				
Time, hr	Serum Conc. off Hemodialysis, µg/ml	Arterial Serum Conc. On Hemodialysis, µg/ml	Venous Serum Conc. On Hemodialysis, µg/ml	Conc. in Spot Sample of Hemodialysis Fluid, µg/ml
0.17 hr	118 ± 17[c]	-	-	-
1	72 ± 7	59 ± 7	58 ± 11[b]	0 ± 0
2	62 ± 7	45 ± 4	34 ± 3	4.4 ± 0.7
3	-	34 ± 3	26 ± 2	2.9 ± 0.3
4	-	27 ± 3	21 ± 2	2.0 ± 0.3
5	-	20 ± 3	17 ± 2	1.5 ± 0.2
6	38 ± 4	-	-	-
12	20 ± 4	-	-	-
24	8.5 ± 5.0	-	-	-
48	2.5 ± 2.3	-	-	-
AUC, µg hr/ml	876 ± 223	-	-	-

[a] All values are mean ± SEM for 6 patients. A Gambro Lundia Plate 11.5-micron hemodialysis machine with a 1-m² effective surface area, blood flow rate 200-250 ml/min, and dialysate flow rate approximately 500 ml/min, was used for all 6 patients. Urinary creatinine clearance ranged from [redacted] ml/min (mean 2.2 ml/min).

[b] Hemodialysis session (~4 hours) began 1 hour after I.V. infusions of aztreonam

[c] Not recorded serum conc. at 0.33 and 0.5 hrs

CAPD Patients[a]				
	Intravenous Dose		Intraperitoneal Dose	
Time, hr	Serum Conc., µg/ml	Dialysate Conc., µg/ml	Serum Conc., µg/ml	Dialysate Conc., µg/ml
0.17	204 ± 58	4.3 ± 1.6	5.8 ± 1.5	274 ± 58
1	64 ± 3	13 ± 3	16 ± 3	260 ± 19
6	33 ± 1	21 ± 3	30 ± 3	105 ± 13
12	18 ± 2	8.4 ± 0.8	18 ± 4	12 ± 2
24	5.9 ± 1.8	2.7 ± 0.6	4.0 ± 0.8	2.0 ± 0.3
48	0.8 ± 0.7	0.3 ± 0.2	0.4 ± 0.4	0.2 ± 0.1
AUC, µg hr/ml	808 ± 84	274 ± 30	454 ± 56	-

[a] All values are mean ± SEM for 6 patients. CAPD involved the use of 2000-ml

Investigators: W.K. Bolton, M.D., U. of Virginia, Charlottesville Va.

Formulation: aztreonam/L-arginine (1.0/0.78)

TABLE 26

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AND PERITONEAL DIALYSIS FLUID AFTER A 1-GRAM INTRAVENOUS DOSE IN PATIENTS UNDERGOING CONTINUOUS AMBULATORY PERITONEAL DIALYSIS

Time After Dosing, hr	Serum Conc.,[a] µg/ml	Dialysate Conc.,[a] µg/ml
0	0.0 ± 0.0	0.0 ± 0.0
0.17	204 ± 58	4.26 ± 1.62
0.33	93.7 ± 8.0	7.15 ± 2.57
0.5	77.9 ± 8.9	7.20 ± 1.98
1	64.5 ± 3.3	12.71 ± 2.96
2	53.5 ± 5.0	16.56 ± 3.25
3	-	17.76 ± 3.33
4	-	19.25 ± 2.95
6	32.8 ± 1.4	20.78 ± 2.92
12	17.7 ± 1.5	8.44 ± 0.77
18	-	4.82 ± 0.56
24	5.90 ± 1.83	2.70 ± 0.62
30	-	1.52 ± 0.46
36	2.40 ± 1.91[r]	0.87 ± 0.48
48	0.82 ± 0.71	0.27 ± 0.23

[a] Values are mean ± S.E.M. for 6 subjects.

[r] 36-hr serum concentration for Subject 12 was 14.0 µg/ml, which was over 4 times the 24-hr value for this subject. The 36-hr value was excluded from the calculated mean value at 36 hr.

TABLE 27

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AND PERITONEAL DIALYSIS FLUID AFTER A 1-GRAM INTRAPERITONEAL DOSE IN PATIENTS UNDERGOING CONTINUOUS AMBULATORY PERITONEAL DIALYSIS

Time After Dosing, hr	Serum Conc.,[a] µg/ml	Dialysate Conc.,[a] µg/ml
0	0.0 ± 0.0	0.0 ± 0.0
0.17	3.80 ± 1.51	274 ± 58
0.33	8.14 ± 2.02	3[illegible] ± 22
0.5	10.94 ± 2.80	304 ± 27
1	15.86 ± 3.38	260 ± 19
2	23.6 ± 3.9	200 ± 15
3	-	166.7 ± 11.4
4	-	140.8 ± 10.3
6	30.0 ± 3.3	104.8 ± 13.4
12	18.2 ± 4.1	11.72 ± 1.50
18	-	4.28 ± 0.47
24	3.96 ± 0.82	1.99 ± 0.29
30	-	1.02 ± 0.35
36	1.07 ± 0.63	0.50 ± 0.26
48	0.43 ± 0.38	0.17 ± 0.14

[a] Values are mean ± S.E.M. for 6 subjects.

that would be bactericidal to commonly encountered members of *Enterobacteriaceae* and *Pseudomonas aeruginosa* strains. Aztreonam underwent hemodialysis (Figure 5) and peritoneal dialysis (Figure 6). Although the monobactam given intravenously displayed good penetration into peritoneal fluid (Figure 6a), intraperitoneal administration (Figure 6b) would be preferable if it were necessary to achieve high antibiotic concentrations in peritoneal fluid.

The serum pharmacokinetics of aztreonam could be described by an open, linear, two-compartment model, with pharmacokinetic parameters shown in the following table.

Parameter[a]	Hemodialysis Patients (N=6)		CAPD Patients (N=6)
	Off Dialysis IV Dose	On Dialysis IV Dose	IV Dose[b]
Distribution			
Extent			
V_1, liters/kg	0.10 ± 0.02	-	0.06 ± 0.07
V^{ss}, liters/kg	0.21 ± 0.02	-	0.16 ± 0.0[illegible]
V^{ss}_{area}, liters/kg	0.22 ± 0.02	-	0.19 ± 0.01
Rate			
$t_{\frac{1}{2}\alpha}$, hr^{-1}	0.18 ± 0.04	-	0.19 ± 0.06
k_{12}, hr^{-1}	3.90 ± 1.93	-	5.26 ± 2.08
k_{21}, hr^{-1}	1.95 ± 0.35	-	0.93 ± 0.24
Elimination			
Extent			
48-hr urinary excr., Xdose	2.0 ± 0.9	-	1.9 ± 1.4
48-hr CAPD elim., Xdose	-	-	9.7 ± 1.0
4-hr HD elim., Xdose	-	38 ± 4	-
serum clear., ml/min	24.4 ± 4.2	-	23.3 ± 2.5
renal clear., ml/min	0.5 ± 0.3	-	0.5 ± 0.4
non-renal clear., ml/min	23.9 ± 4.0	-	*
HD clear.,[c] ml/min, and	-	43.2 ± 1.6	-
% of HD clear. of urea	-	28 ± 2	-
CAPD clear., ml/min, and	-	-	2.1 ± 0.3
% of CAPD clear. of urea	-	-	32 ± 4
non-renal, non-CAPD clear., ml/min			21.3 ± 2.4
Rate			
$t_{\frac{1}{2}\beta}$, hr^{-1}	7.94 ± 2.51	2.67 ± 0.29[d]	7.08 ± 1.43
k_{10}, hr^{-1}	0.31 ± 0.09	-	1.57 ± 0.71

[a] All values are mean ± SEM.

[b] The extent of absorption of an intraperitoneal dose was 59 ± 10% of dose, based on serum AUC after IP dose/serum AUC after IV dose, and was 73 ± 2% of dose, based on (dose - amount recovered from abdomen after 6 hr)/dose. *

[c] Based on total amount of aztreonam recovered in dialysate/serum AUC.

[d] Based on aztreonam concentrations in arterial serum.

[e] Composed of clearance by CAPD (2.1 ± 0.3 ml/min), and non-renal/non-CAPD (21.3 ± 2.4 ml/min) processes.

The bioavailability of intraperitoneally administered aztreonam was 59%, based on the ratio of serum AUC after intraperitoneal dosing to serum AUC after intravenous dosing, and was 73%, based on comparison of the amounts of drug administered intraperitoneally and subsequently recovered in fluid drained at the end of the 6-hour dwell time. A standard 4-hour hemodialysis treatment could remove 27 to 58% (mean 38%) of a prior 1000-mg dose, whereas CAPD could remove 6 to 12% (mean 9.7%) of a 1000-mg intravenous dose in 48 hours. The clearance of aztreonam by either method of dialysis was about 30% of the simultaneously measured clearance of urea by the corresponding method of dialysis. Clearance of aztreonam by hemodialysis was about 2 times serum clearance off hemodialysis, while clearance by CAPD was only approximately 10% of serum clearance. Consequently, clearance of aztreonam by hemodialysis was about 20 times that by CAPD. The elimination half-life of aztreonam given intravenously to hemodialysis patients off dialysis and given intravenously to CAPD patients was similar. Although most patients with severe renal insufficiency had elimination half-lives (off hemodialysis or during CAPD) of 4.8 to 7.0 hours, two patients who had both lower extremities amputated had values in the 14- to 20-hour range. However, the explanation for this finding was not apparent. *

Aztreonam dosage regimens should be adjusted in patients with severe renal insufficiency. A loading dose equal to the standard dose, followed at the standard dose interval by one-fourth the standard dose intravenously, is appropriate for both hemodialysis patients off dialysis and CAPD patients. Hemodialysis patients should receive one-eighth the standard dose after a standard dialysis treatment, to compensate for drug cleared by dialysis. High aztreonam levels in peritoneal dialysis fluid could be achieved by 500-mg q6h intraperitoneal dosing in CAPD patients, preceded by a 1000-mg intravenous loading dose. *

The PK parameters for the intravenous dose (i.e. off dialysis) are similar to those determined in Study Protocol # 18,554-8 for the subjects with creatinine clearance <10 ml/min.

From Study Protocols # 18,554-24 and # 18,554-25 the sponsor recommended that following hemodialysis, aztreonam should be replaced for the amount of drug lost by dialysis. In each of these studies different hemodialysis equipment was used along with different flowrate. However, the amounts of drug recommended for replacement were similar

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AFTER A
1-GRAM 2-MINUTE INFUSION IN PATIENTS
ON AND OFF HEMODIALYSIS

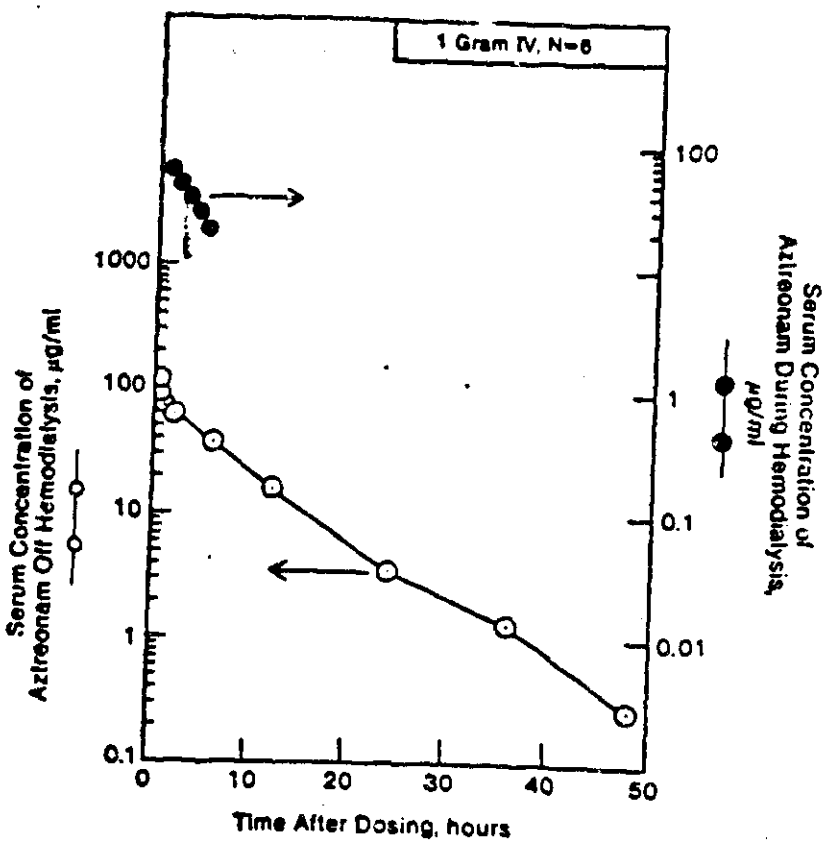


FIGURE 2

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AND DIALYSATE
AFTER A 1-GRAM 2-MINUTE INFUSION
IN PATIENTS UNDERGOING CONTINUOUS
(DWELL TIME 6 HOURS) AMBULATORY PERITONEAL DIALYSIS

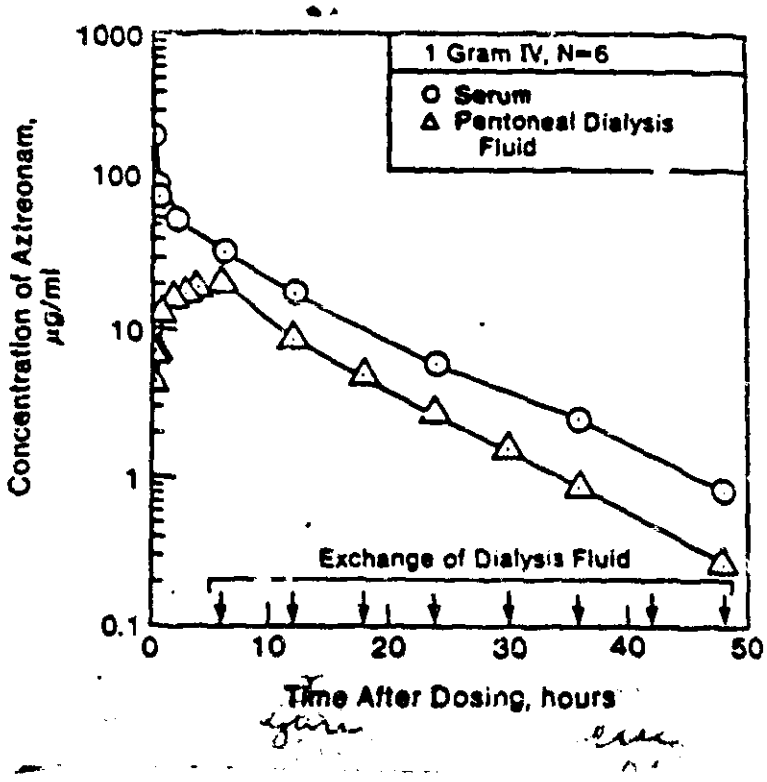
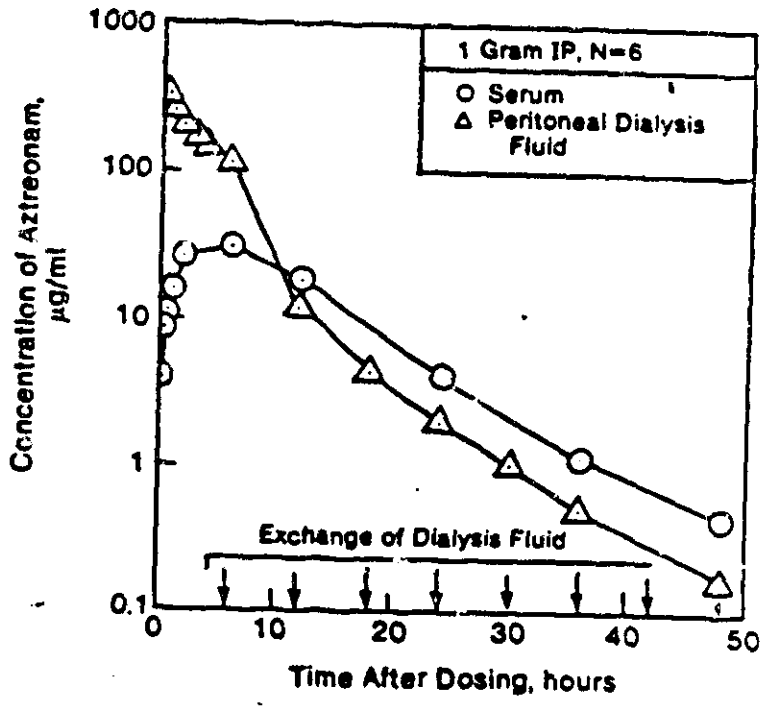


FIGURE 3

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AND DIALYSATE
AFTER A 1-GRAM INTRAPERITONEAL DOSE
IN PATIENTS UNDERGOING CONTINUOUS (DWELL TIME
6 HOURS) AMBULATORY PERITONEAL DIALYSIS



Investigators: M. A. Apicella, M.D. and W. J. Jusko, Ph.D., State U. of New York, Buffalo, N. Y.

Pharmacokinetics of aztreonam in patients with lower respiratory tract infections (Addendum A to Protocol 18554-11) or serious urinary tract infections (Addendum B to Protocol 18554-14)

Pharmacokinetic profiles of aztreonam were determined in 1 patient with a lower respiratory tract infection and 6 patients with serious urinary tract infections on the first or second and last days of aztreonam treatment (1000 or 2000 mg aztreonam intravenously q8h for 7 to 11 days). Two other patients with urinary tract infections left the study early due to difficulty in obtaining venous access. The evaluable patients had creatinine clearances from 16 to 127 ml/(min x 1.73 sq m). Multiple serum and urine specimens were obtained from each patient and analyzed by the investigators' laboratory using high-pressure liquid chromatography methods.

The 4 male and 3 female patients (5 Caucasian, 2 Negro) who completed this study had ages ranging from 44 to 76 years (mean, 63 years) and body weights ranging from 54.0 to 86.4 kg (mean, 71.4 kg).

Mean (±SEM) values for peak (C_{max}) and trough (C_{min}) serum concentrations, area under the serum concentration-time curve (AUC), elimination half-life ($t_{1/2}$), renal excretion, steady-state volume of distribution (V_{ss}), and serum, renal, and non-renal clearances on both study days are shown in the next table.

Parameter	Day 1 or 2	Last Day
C_{max}, µg/ml[a]	127.0 ± 22.3	79.5 ± 7.4 → 37% decrease
C_{min}, µg/ml[a]	24.6 ± 9.1	12.0 ± 4.6 → 51% "
AUC, µg hr/ml[a]	408 ± 105	240 ± 39 → 41% "
$t_{1/2}$, hr	3.83 ± 0.83	3.55 ± 1.00
serum clear., ml/hr/kg	50.1 ± 11.9	65.8 ± 10.2 → 31% increase
renal clear., ml/hr/kg	31.4 ± 8.2	38.7 ± 6.6 → 23% "
non-renal clear., ml/hr/kg	18.7 ± 3.8	27.1 ± 5.2 → 45% "
V_{ss}, liters/kg	0.15 ± 0.02	0.16 ± 0.02
creatinine clearance ml/min/1.73 sq m	51 ± 16	70 ± 14

[a]Based on 1000-mg doses. C_{max}, C_{min}, and AUC were normalized to 1000-mg doses for the one patient receiving 2000-mg doses.

The investigator indicated that the increase in non-renal clearance of aztreonam may have been related to improved hepatic function as indicated by improvement in liver function test values in most patients.

There was a significant improvement in the elimination of aztreonam from the first or second day to the last day of therapy due to improvement in renal function (related in most cases to reversal of dehydration). Serum protein binding averaged 30%, approximately half that previously found in healthy volunteers, perhaps due to undefined factors relating to infection, uremia, drug interactions, or to differences in technique used in measuring binding.

This study confirmed in infected patients the following previously-reported findings in healthy young volunteers and uninfected patients with renal insufficiency: 1) aztreonam half-life is approximately 2 hours when renal function is normal, 2) aztreonam is normally eliminated unchanged in the urine, 3) there is no appreciable accumulation of aztreonam during q8h multiple dosing, 4) potentially therapeutic serum and urinary levels of aztreonam can be achieved with multiple intravenous doses of 1000 or 2000 mg, 5) renal insufficiency can markedly reduce the serum clearance and prolong the elimination half-life of aztreonam, and 6) aztreonam dosages in patients with mild, moderate, or severe renal insufficiency (defined as creatinine clearances of 30 to 80, 10 to 30, and less than 10 ml/min) can be one-half, one-third, and one-fourth the standard aztreonam dose at the standard dose interval (based upon correlations of serum clearance of aztreonam with creatinine clearance).

Note: This study gives insight into the effect of renal function as related to proposed package ins dose adjustments using multiple dose drug serum clearance and creatinine clearance data (figure 2). However it is important to note that

FIGURE 1

SERUM CONCENTRATION-TIME PROFILE ON THE FIRST AND LAST DAYS OF AZTREONAM TREATMENT IN PATIENT 11, PROTOCOL 18554-14

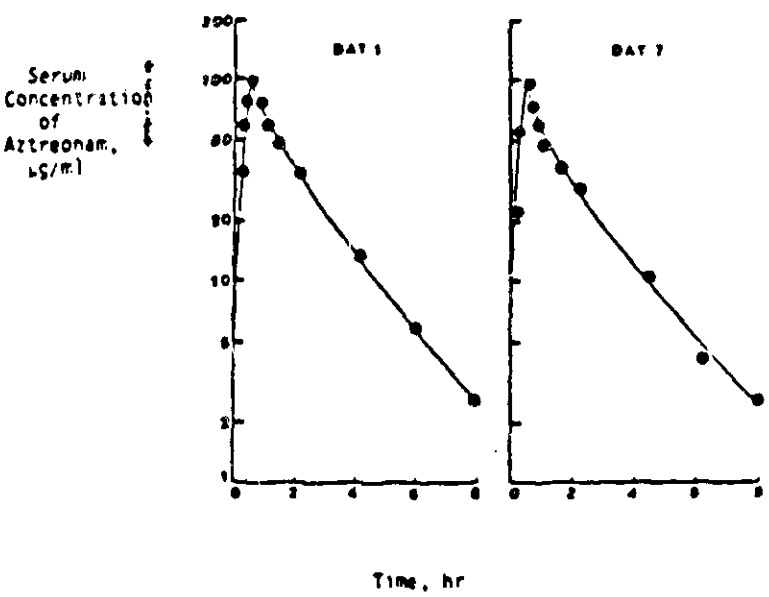
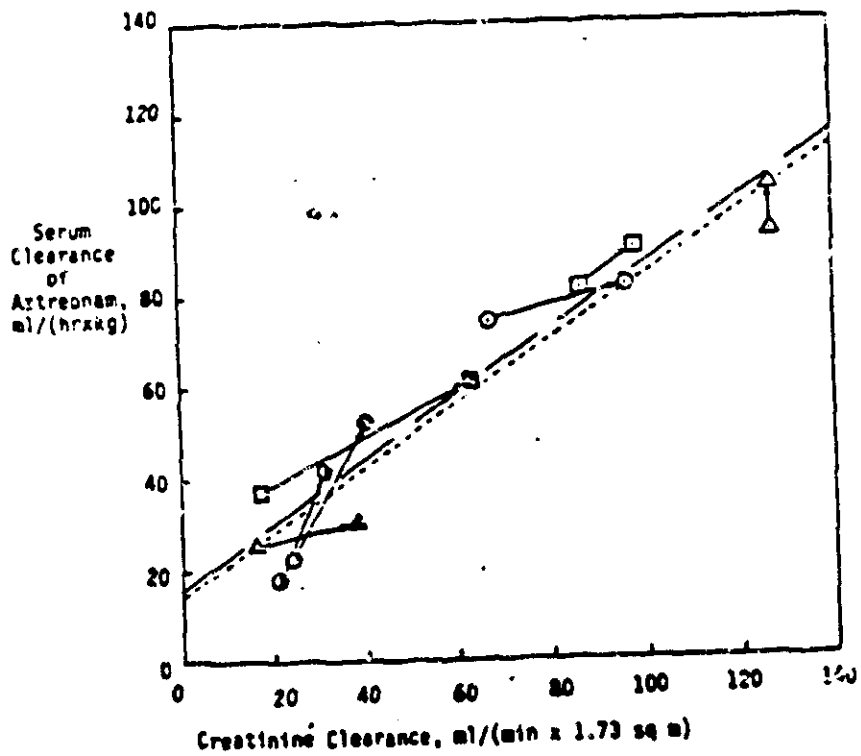


FIGURE 2

VARIATION OF SERUM CLEARANCE OF AZTREONAM WITH CREATININE CLEARANCE



Patient No. 4 ○, 7 □, 8 ○, 11 △, 12 ●, 14 ■, 16 ▲.

Regression Lines: Day 1 or 2 Data: CL = 0.691 CrCL + 14.7 ——
Day 7 to 11 Data: CL = 0.710 CrCL + 15.8 .....

It is apparent t
there was no significant difference between these two relati
suggesting that the infectious state does not, in itself, al
functional relationships of serum aztreonam clearance with
creatinine clearance.

Investigator: F.K. Sattler, M.D., Hershey Medical Center, Penn State University, Hershey, PA

Pharmacokinetics of aztreonam in patients with urinary tract infections and renal insufficiency (Addenda A to Protocols 18554-27 and -31)

Serum trough concentrations (levels in serum obtained just prior to a scheduled dose) of aztreonam and SQ 26,992 were measured daily in 9 patients (2 enrolled in Protocol 18554-27 and 7 enrolled in Protocol 18554-31) with renal insufficiency. These patients were receiving 500, 1000, or 2000 mg aztreonam intravenously q8h x 5 to 10 days for treatment of serious urinary tract infections. One patient received hemodialysis and provided additional blood specimens before and after hemodialysis. Serum concentrations of aztreonam and SQ 26,992 (the major metabolite of aztreonam resulting from hydrolytic opening of the beta-lactam ring) were measured in all patients by high-pressure liquid chromatography.

The 5 male and 4 female Caucasian patients enrolled in these addenda studies had ages ranging from 56 to 83 years (mean, 72 years) and body weights from 49.5 to 81.7 kg (mean 67.7 kg).

Aztreonam trough levels were generally stable in the 20 to 70 µg/ml range during treatment. Such levels would be potentially therapeutic for commonly encountered *Enterobacteriaceae* and *Pseudomonas aeruginosa*. SQ 26,992 concentrations tended to increase to steady-state levels during therapy in proportion to the degree of renal insufficiency. Initial serum levels of SQ 26,992 were in the 1.9 to 5.0 µg/ml range, and Day 5 or 6 levels were in the 3.0 to 29.2 µg/ml range. The ratio of Day 5 or 6 serum SQ 26,992 concentration to the initial serum level (before the third or fourth dose on Day 1 or 2) was near unity in patients with creatinine clearances in the 25 to 40 ml/min range, and was between 3.5 and 11.3 in patients with creatinine clearances below 25 ml/min. Aztreonam and SQ 26,992 underwent hemodialysis; nevertheless, the highest serum levels of SQ 26,992 were measured in a patient being supported on chronic hemodialysis.

Aztreonam was tolerated well by patients with serious urinary tract infections. A possible drug-related adverse effect in 1 of 9 patients consisted of mildly elevated serum transaminases. Thus, the accumulation of SQ 26,992 had no clinically important effects.

TABLE 1

BIOGRAPHIC DATA AND DOSAGE REGIMENS

Addendum Patient No. (Initials)	Main Protocol Pt. No. 18554-		Age	Sex	Height (cm)	Wt. (kg)	Aztreonam Dose (IV)
1	-31	5	56	M	NR	81.7	1gm q8h x 5d
2	-31	7	75	F	162.6	70.5	0.5gm q8h x 6d
3	-27	51	75	F	169.5	60.4	2gm q8h x 10d
4	-27	52	83	F	152.0	49.5	2gm q8h x 8d
5	-31	9	66	F	171.0	80.4	1gm loading, 0.5gm q8h x 6d
6	-31	10	70	M	174.0	83.4	1gm loading, 0.5gm q8h x 7d
7	-31	12	81	M	NR	66.5	2gm loading, 1gm q8h x 6d
8	-31	14	80	M	NR	60.0	1gm q8h x 6d
9	-31	8	62	M	NR	76.6	1gm loading, 0.5gm q8h x 6d
Mean (Range)			72 (56-83)			67.7 (49.5-81.7)	

* Calculated using nomogram of Sinkule et al, 1979.

TABLE 1 (cont.)

SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992

Add. Pt. Nos.	4		5		6	
Treatment Day	AZ	SQ 26,992	AZ	SQ 26,992	AZ	SQ 26,992
1	43.0*	2.56*	NR	NR	32.1*	2.31*
2	43.6	5.26	43.0*	6.04*		
3	40.5	8.33	36.7	8.94	31.8	6.43
4	40.8	4.90	41.0	13.6	28.8	7.83
5	38.9*	4.63*	45.5	17.9	32.1	8.74
6	40.3	5.24	44.4*	20.8*	27.5*	8.01*
7	42.0	4.98	NR	NR	NR	NR
8	NR	NR				
9						
10						
11						
12						
13						
14						
*Ratio of Serum Conc. Day 5 or 6 to Day 1 or 2	0.81	1.02	1.03	4.12	0.86	3.47

TABLE 2

SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992

Add. Pt. Nos.	1		2		3	
Treatment Day	AZ	SQ 26,992	AZ	SQ 26,992	AZ	SQ 26,992
1	30.3*	1.93*	NR	NR	27.1*	2.59*
2	68.2	8.57	28.1*	2.32*	43.8	3.63
3	57.0	8.42	37.1	8.84	24.6	3.15
4	68.5	16.2	26.0	12.8	19.5	2.87
5	51.7*	14.9*	21.8	14.8	27.7*	3.99*
6	NR	NR	18.5*	13.9*	NR	NR
7			NR	NR	34.9	3.52
8					20.6	2.94
9					18.2	4.25
10					9.1	1.89
11					NR	NR
12						
13						
14						
*Ratio of Serum Conc. Day 5 or 6 to Day 1 or 2	1.74	1.72	0.66	5.99	1.02	0.87

*Day 1 was day of first dose of aztreonam. The first serum specimen was obtained prior to the third or fourth dose, which fell on Day 1 or 2 for various patients.

TABLE 2 (cont.)

SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992

Add. Pt. Nos.	7		8		9	
Treatment Day	AZ	SQ 26,992	AZ	SQ 26,992	AZ	SQ 26,992
1	NR	NR	14.2	NR	NR	NR
2	86.7*	3.92*	17.1*	2.23*	31.3*	2.66*
3	89.3	4.43	46.8	8.77	41.3	3.85
4	83.0	8.64	17.4	3.98	65.4	9.77
5	31.3	3.28	19.6	3.27	80.9	10.9
6	27.1*	3.14*	24.3*	3.31*	78.2*	10.5*
7	35.3	4.31	17.7	3.76	71.8	10.2
8	48.6	4.57	15.6	3.48	64.8	10.2
9	NR	NR	NR	NR	NR	NR
10						
11						
12						
13						
14						
*Ratio of Serum Conc. Day 5 or 6 to Day 1 or 2	0.83	1.01	1.43	1.48	2.04	11.3

This study suggests that the major metabolite of aztreonam does not accumulate to clinically significant levels in patients with renal insufficiency.

TABLE 6

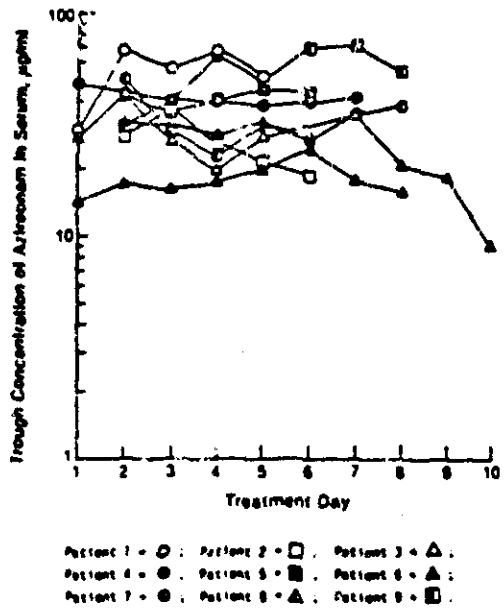
EFFECT OF HEMODIALYSIS ON SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992 IN PATIENT 7

Treatment Day	Pre dose AZ	Pre dose SQ 26,992	Post dose AZ	Post dose SQ 26,992	Pre hemodialysis AZ	Pre hemodialysis SQ 26,992	Post hemodialysis AZ	Post hemodialysis SQ 26,992
2	31.1	2.69	38.5	3.13	31.0	2.88	11.2	2.52
3	41.4	1.47	71.1	3.31	.	.	.	.
4	55.4	8.27	55.5	9.97	29.8[b]	3.74[b]	24.2[b]	10.2[b]
5	38.8	10.9	62.4	19.1	.	.	.	.
6	28.2	29.2	38.2	29.8	.	.	.	.
7	31.8	38.2	38.8	41.4	68.6	38.1	21.4	14.1
8	14.6[c]	25.2[c]	42.6[c]	23.8[c]	.	.	.	.

[a] Data from the Squibb Institute.
[b] In view of the well-documented clearance of aztreonam during hemodialysis (Protocol 18554-75), it seems probable that the serum specimens sent to the Squibb Institute for Day 6 pre and post hemodialysis were inadvertently mislabeled. [order switched]
[c] In view of the repeated demonstration on Days 2 through 7 that post-dose aztreonam levels exceeded pre-dose levels, it seems probable that the serum specimens sent to the Squibb Institute for Day 8 were inadvertently mislabeled [order switched]

FIGURE 1

INDIVIDUAL TROUGH CONCENTRATIONS OF AZTREONAM IN SERUM OF PATIENTS RECEIVING 0.5, 1, OR 2 GRAMS OF AZTREONAM INTRAVENOUSLY Q8h AS THERAPY FOR SERIOUS URINARY TRACT INFECTIONS

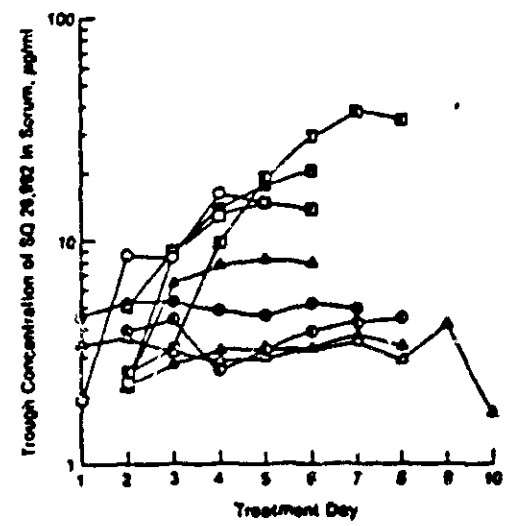


Patient 1 = ○ ; Patient 2 = □ ; Patient 3 = △ ;
Patient 4 = ● ; Patient 5 = ■ ; Patient 6 = ▲ ;
Patient 7 = ◐ ; Patient 8 = ◭ ; Patient 9 = ◧ .

Appendix A to
Protocols 18554-27
and 18554-31

FIGURE 2

INDIVIDUAL TROUGH CONCENTRATIONS OF SQ 26,992 IN THE SERUM OF PATIENTS RECEIVING 0.5, 1, OR 2 GRAMS OF AZTREONAM INTRAVENOUSLY Q8h AS THERAPY FOR SERIOUS URINARY TRACT INFECTIONS



Patient 1 = ○ ; Patient 2 = □ ; Patient 3 = △ ;
Patient 4 = ● ; Patient 5 = ■ ; Patient 6 = ▲ ;
Patient 7 = ◐ ; Patient 8 = ◭ ; Patient 9 = ◧ .

See section IV of this review where this study is discussed in further detail.

Renal tubular handling of aztreonam in healthy subjects (Protocol 18554-6)

Aztreonam was administered by an intravenous loading dose of 1200 mg over 2 minutes followed by a continuous infusion of 500 mg/hr for 4 hours to 6 healthy male volunteers with and without co-administration of probenecid (1 gram po bid for 2 days prior to aztreonam infusion and during the day of infusion). To assess glomerular filtration, each subject also received inulin as an intravenous loading dose of 30 mg/kg followed immediately by a continuous infusion of 35 mg/min for 4.75 hours. One additional subject was dropped from the study for failing to follow the dosage regimen for probenecid. To assess the safety of aztreonam, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and at frequent intervals during the study.

The 6 male volunteers (5 Caucasian, 1 Negro) completing this study ranged in age from 20 to 32 years (mean, 25 years) and in body weight from 61.2 to 75.6 kg (mean, 71.8 kg).

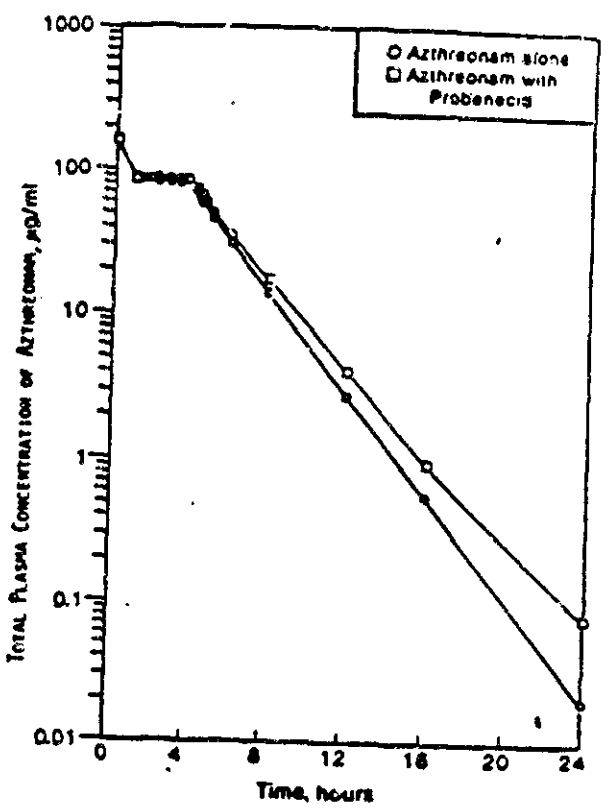
Aztreonam was tolerated well by healthy male subjects. Possible drug-related side effects occurred in 3 subjects. One subject experienced fever (100.4°F), headache, and malaise 24 hours after receiving aztreonam with probenecid, during the second leg of the study. A second subject experienced an erythematous, pruritic rash 25 hours after receiving aztreonam with probenecid, also during the second leg of the study. A third subject reported mild taste alterations during both 2-minute infusions.

The pharmacokinetic profile of aztreonam was assessed by measuring aztreonam concentrations in multiple plasma and urine samples during each dosage regimen. Binding of aztreonam to plasma proteins was assessed by preparation of protein-free filtrate of plasma by ultracentrifugation. Microbiological methods were used to assay aztreonam in each sample. Inulin levels were also determined in plasma and urine by standard methods.

Mean serum concentrations of aztreonam in the presence and absence of probenecid are shown in Figure 8a. The following table summarizes various parameters (mean ± SEM) for aztreonam in the presence and absence of probenecid.

FIGURE 1

MEAN TOTAL PLASMA AZTHREONAM CONCENTRATIONS VS. TIME IN THE PRESENCE AND ABSENCE OF PROBENECID



Parameter	Aztreonam Alone	Aztreonam Plus Probenecid	P
Steady-State (2 to 4 hours)			
Total Plasma Concentration, ug/ml	81.7 ± 3.4	86.0 ± 2.2	<0.05
Free Plasma Concentration, ug/ml	33.1 ± 2.2	41.5 ± 3.0	<0.01
Plasma Protein Binding, %	59.6 ± 1.4	52.1 ± 2.2	<0.05
Plasma Clearance of Free Drug, ml/min/kg	3.57 ± 0.17	2.86 ± 0.14	<0.01
Renal Clearance of Free Drug, ml/min/kg	2.81 ± 0.13	2.02 ± 0.12	<0.01
Glomerular Filtration Rate, ml/min/kg	1.52 ± 0.07	1.43 ± 0.12	NS
Tubular Secretion, % of renal clearance	45.8 ± 1.9	28.0 ± 3.4	<0.05
Non-renal Clearance of Free Drug, ml/min/kg	0.76 ± 0.21	0.84 ± 0.10	NS
Steady-State Volume of Distribution, liters/kg[a]	0.42 ± 0.02	0.35 ± 0.02	<0.01
Elimination-Phase (4 to 48 hours)			
ß-Phase half-life, hr[b]	1.76 ± 0.03	1.95 ± 0.06	<0.01
Cumulative Urinary Excretion, % dose[c]	71.9 ± 5.4	63.3 ± 3.2	<0.05

[a] Based on free drug concentration in plasma.
[b] Based on total drug concentration in plasma.
[c] 0-48 hours.

These results indicated that aztreonam was excreted in the urine by both glomerular filtration and tubular secretion in essentially equal proportions. Probenecid reduced plasma clearance by suppressing renal tubular secretion without significantly altering glomerular filtration rate (Figure 8b). Probenecid also increased total and free aztreonam levels and aztreonam half-life in plasma, while reducing plasma protein binding and apparent steady-state volume of distribution.

Continuous infusion of 500 mg/hr of aztreonam achieved plasma levels comparable to published values for penicillins and cephalosporins administered by the same regimen. This regimen would produce plasma levels of aztreonam that would exceed the MIC_{90} for most aerobic gram-negative bacteria, including *Pseudomonas aeruginosa*.

This study demonstrated that probenecid does not significantly effect the pharmacokinetics of aztreonam such to warrant major concerns of drug dose adjustment with concomitant probenecid administration.

SECTION:
Division of Medical Affairs

SUBJECT: SQ 18,778 (Aztreonam)

TITLE:
Report on Safety and Pharmacokinetic Study of Intravenous Aztreonam and Oral Furosemide in Healthy Subjects.

Study Protocol No. 18,544-19

AUTHORS:
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INVESTIGATORS:
A. A. Sugerman, M.D., Princeton Medical Center; T.B. Platt, Ph.D., The Squibb Institute

ABSTRACT

Aztreonam was administered as a single 1000-mg intravenous infusion over 2 minutes, alone or preceded by an 80-mg oral dose of furosemide, to 9 healthy male volunteers according to a two-way crossover design with a 7-day washout period between drug treatments. An additional subject did not complete the entire study due to unsuccessful intravenous administration of the second aztreonam dose. To assess the safety of aztreonam alone and in combination with furosemide, physical and electrocardiographic examinations, monitoring of vital signs, and clinical laboratory tests were conducted before and at frequent intervals after each drug treatment.

Aztreonam alone and in combination with furosemide was tolerated well by healthy male subjects. Possible drug-related adverse reactions consisted of transient taste alteration during the intravenous infusion (1/9 subjects), transient elevation in SGPT (1/9 subjects), and transient pyuria (white cells in the urine) and cylindruria (epithelial cell casts in the urine) (1/9 subjects).

Results of special renal function tests considered to be highly sensitive indicators of even subclinical renal injury suggest that the combination of aztreonam with furosemide is as safe as aztreonam alone, for healthy male subjects receiving single drug treatments.

The pharmacokinetic profile of aztreonam was assessed by measuring aztreonam concentrations in multiple samples of serum, urine, and saliva after administration of aztreonam. Maximum serum concentrations (C_{max}) and areas under the serum concentration-time curves (AUC) for aztreonam given alone and preceded by an 80-mg oral dose of furosemide are shown in Table I.

TABLE I

PARAMETER[a]	AZTREONAM ALONE	AZTREONAM PLUS FUROSEMIDE	p[b]
C_{max}, µg/ml	100.7 ± 2.5	88.6 ± 8.9	NS
$AUC_{0-\infty}$, µg × hr/ml	147.6 ± 7.2	163.1 ± 8.9	NS

[a] Values are arithmetic mean ± S.E.M. for 9 subjects.

[b] Based upon analysis of variance for the crossover design using log-transformed data.

The serum pharmacokinetics of intravenously administered aztreonam were analyzed by model-independent methods, which could be applied uniformly to data from all subjects, regardless of individual differences in pharmacokinetic profiles. A detailed summary of the pharmacokinetics of aztreonam is given in Table II.

TABLE II

PARAMETER[a]	AZTREONAM ALONE	AZTREONAM PLUS FUROSEMIDE	p[b]
Distribution			
Extent			
V_{ss}, liters/kg	0.19 ± 0.01	0.19 ± 0.01	NS
Elimination			
Extent			
urine, % of dose, 0 to 24 hr	61.1 ± 1.7	59.9 ± 2.0	NS
serum clear., ml/(min kg)	1.44 ± 0.07	1.38 ± 0.06	NS
renal clear., ml/(min kg)	0.88 ± 0.05	0.77 ± 0.02	<0.05
nonrenal clear., ml/(min kg)	0.56 ± 0.04	0.63 ± 0.06	NS
Rate			
$t_{\frac{1}{2}}$, hr	1.72 ± 0.08	1.92 ± 0.19	NS
MRT, hr	2.23 ± 0.09	2.57 ± 0.22	<0.05

[a] Values are arithmetic mean ± S.E.M. for 9 subjects.

[b] Based upon analysis of variance for the crossover design using log-transformed data.

Furosemide increased the serum half-life and mean residence time (MRT) of aztreonam, and reduced the serum and renal clearance of aztreonam, without altering the apparent volume of distribution at steady state or nonrenal elimination. Although these effects were relatively minor in magnitude, they could be explained by the likely possibility that furosemide and aztreonam compete for the same organic anion transport site on the renal tubular cell. In the presence or absence of furosemide, the primary route of elimination of aztreonam was renal excretion of unchanged drug. Urinary excretion of aztreonam was essentially complete by 8 hours.

Salivary levels of aztreonam were less than 1% of concurrently measured serum levels after a single 1000-mg dose of aztreonam. The assessment of salivary levels of aztreonam would appear not to have any practical value in therapeutic monitoring of this monobactam antibiotic.

SERUM CONCENTRATIONS (MEAN ± SEM, µgm/ml) OF AZTREONAM AFTER A 2-MIN INTRAVENOUS INFUSION OF 1000 MG OF AZTREONAM ALONE AND PRECEDED BY AN 80-MG ORAL DOSE OF FUROSEMIDE IN NINE HEALTHY MALE VOLUNTEERS

TIME AFTER INFUSION HR	AZTREONAM ALONE[a]	AZTREONAM PLUS FUROSEMIDE
PRE	0.00	0.00
0.08	100.7 ± 2.5	87.8 ± 10.4
0.17	78.5 ± 2.6	70.6 ± 7.0
0.33	62.1 ± 2.6	55.0 ± 4.9
0.50	51.1 ± 1.3	50.4 ± 4.4
1.0	34.7 ± 1.6	37.7 ± 2.7
1.5	27.3 ± 1.3	30.3 ± 1.9
2.0	21.8 ± 1.3	25.0 ± 1.5
3.0	14.4 ± 1.0	18.2 ± 1.3
4.0	10.3 ± 0.8	12.9 ± 1.1
6.0	4.82± 0.48	6.45± 0.82
8.0	2.17± 0.29	3.15± 0.58
12.0	0.43± 0.09	0.67± 0.16
16.0	0.08± 0.03	0.14± 0.05

[a] Mean ± S.E.M. (µgm/ml) salivary concentrations of aztreonam were 0.0 at Pre, 0.16 ± 0.03 at 0.5 hr, 0.08 ± 0.01 at 1.0 hr, and 0.04 ± 0.02 at 3 hr.

FIGURE 1

EFFECT OF FUROSEMIDE ON THE SERUM PHARMACOKINETICS OF AZTREONAM IN NINE HEALTHY MALE VOLUNTEERS

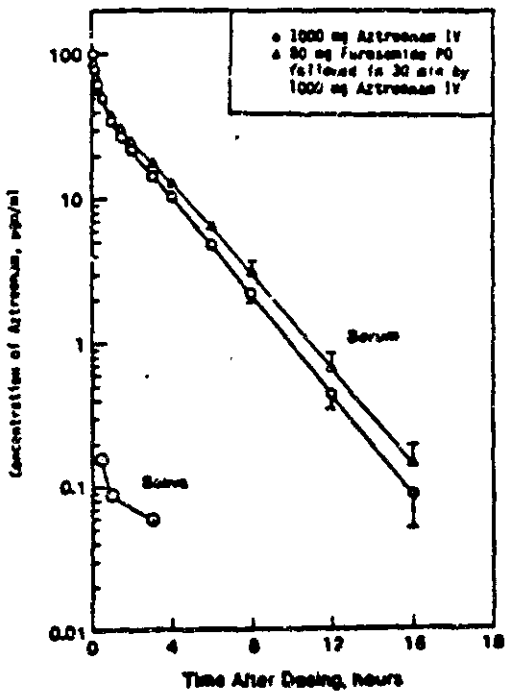
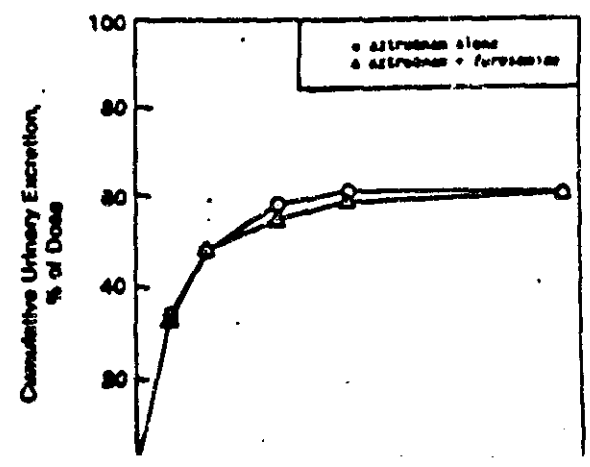


FIGURE 2

RENAL EXCRETION OF AZTREONAM IN THE PRESENCE AND ABSENCE OF FUROSEMIDE IN NINE HEALTHY MALE VOLUNTEERS



Department of Clinical Pharmacology

Division of Medical Affairs

TITLE

Report on the Pharmacokinetic Interaction of Aztreonam (SQ 26,776) with Gentamicin in Healthy Volunteers

Protocol No. 18,554-46

AUTHORS

William A. [illegible]sey, D. Phil., Ellen Hudes, M.S. and Janice Lus, B.S., M.P.H.

INVESTIGATORS A. Arthur Sugerman, M.D., The Medical Center at Princeton, Princeton, New Jersey 08540

→ page 19-31

ABSTRACT

Twelve (12) healthy male volunteers were enrolled in this single-dose, three-way, balanced crossover study. Each subject received, in randomized order, a single 30-minute intravenous infusion of aztreonam (1000 mg), gentamicin (80 mg) or aztreonam (1000 mg) plus gentamicin (80 mg) simultaneously, on each of 3 study days separated by a 7-day washout period. Concentrations of aztreonam and gentamicin were assayed in serum samples collected at the end of the infusion, and 0.25, 0.5, 1, 2, 3, 4, 6, 8, 12 and 24 hours after the infusion. Protein-free filtrates of sera collected 0.25, 1 and 3 hours after the end of the infusion were assayed for aztreonam. The urines collected during the -8 to 0, 0 to 2, 2 to 4, 4 to 8 and 8 to 24 hour periods after infusion were assayed for aztreonam and gentamicin. All subjects received physical examinations, 12-lead electrocardiograms, and a battery of tests of blood and urine chemistry, hematology and serum enzymes reflecting liver function.

Mean serum levels of aztreonam were slightly higher after administration of the monobactam alone than when it was given in combination with gentamicin, but this trend was statistically significant only during the first hour after infusion. These serum differences were reflected in the areas under the serum concentration versus time curves and the maximum serum concentration values. However, although these differences were statistically significant, they were of such a low order of magnitude as to be clinically irrelevant.

FIGURE 1

SERUM LEVELS OF AZTREONAM

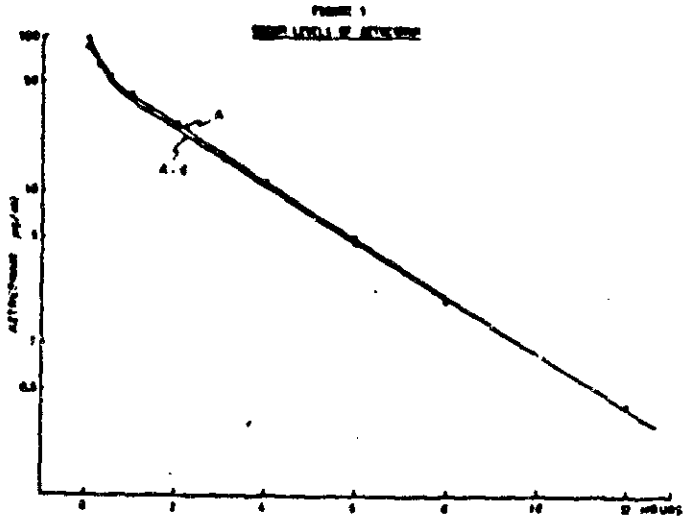
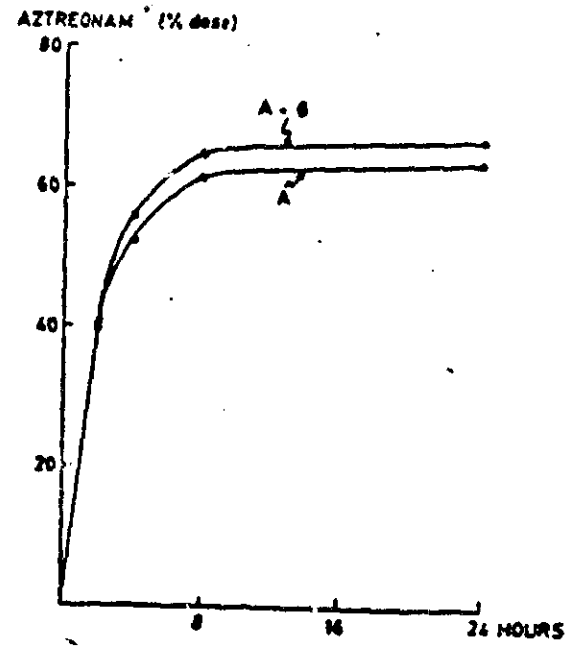


FIGURE 2

CUMULATIVE URINARY EXCRETION OF AZTREONAM



Serum levels of gentamicin were generally lower when the two antibiotics were given in combination than when the aminoglycoside was administered alone, but these differences were statistically significant only at 0.5 and 2 hours. Values for the Cmax, the areas under the curve and the serum half-lives showed no significant differences between monotherapy and the combination.

Urinary excretion of aztreonam reached 63.0 and 66.4 percent of administered dose after single drug or combined treatment, respectively. In the case of gentamicin, the corresponding figures were 68.6 and 75.4 percent, respectively. Only in the 0-4 hour collection was the somewhat greater excretion during combined therapy statistically significant. The table below summarizes the pharmacokinetic data.

	Aztreonam Alone	Gentamicin Alone	Aztreonam plus Gentamicin: Aztreonam	Aztreonam plus Gentamicin: Gentamicin
Cmax (µg/ml)	98.1 ± 5.0	7.0 ± 0.3	85.7 ± 3.6[a]	7.2 ± 0.2
Serum-protein Binding (percent at 0.25 hr)	62.2 ± 2.4	n.d.	60.8 ± 1.5	n.d.
AUC 0-24 µg.hr/ml	362.4 ± 5.4	18.2 ± 0.8	350.6 ± 6.4[b]	24.8 ± 0.8
t½ (hours)	1.62 ± 0.03	3.38 ± 0.24	1.61 ± 0.03	3.00 ± 0.33
Urinary Excretion 0-24 hr (percent of dose)	63.0 ± 3.1	68.6 ± 5.9	66.4 ± 2.7	75.4 ± 2.6

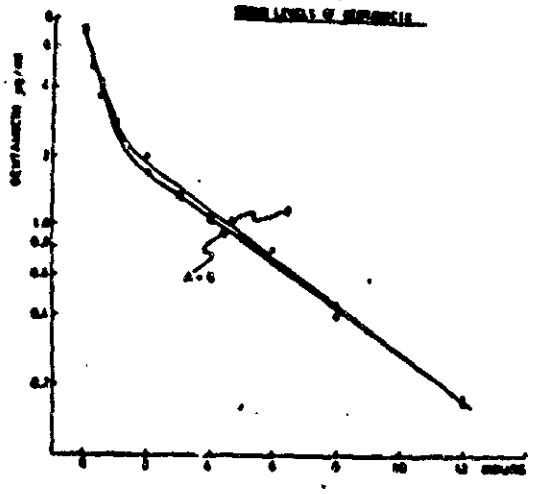
[a] Significantly different from value for aztreonam alone ($p < 0.05$)

[b] Significantly different from value for aztreonam alone ($p < 0.05$)

n.d. not determined.

FIGURE 3

SERUM LEVELS OF GENTAMICIN



Possible adverse reactions to drug that were encountered included transient elevation of serum creatine phosphokinase in one subject after a dose of aztreonam alone, and a mild rise in serum glutamic-pyruvic transaminase after aztreonam and gentamicin in combination. Both parameters returned to normal without further action.

Measurement of the in vitro bactericidal activity of sera indicated that gentamicin neither antagonized nor potentiated the action of aztreonam against Escherichia coli SC 8294.

Department of Clinical Pharmacology

SECTION
Division of Medical Affairs

August 3, 1983

MRL/CCC CODE
MRL-860

SQUIBB CODE OR MRL/CCC CODE
SQ 26,776 (Aztreonam)

TITLE
Report on the Pharmacokinetic Interaction of Aztreonam (SQ 26,776) with Clindamycin in Healthy Volunteers

Study Protocol # 18,554-47

AUTHORS: William A. Creasey, D.Phil, Michelle A. Stern, B. A. and Janice Lux, B.S., M.P.H.

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ABSTRACT

Aged over 30 years

Nine (9) healthy male volunteers were enrolled in this single-dose three-way crossover study. Each subject received, in randomized order, a single 30-minute intravenous infusion of aztreonam (1000 mg), clindamycin (600 mg) or aztreonam (1000 mg) plus clindamycin (600 mg) simultaneously, on each of 3 study days separated by a 7-day washout period. Concentrations of aztreonam and clindamycin were assayed in serum samples collected at the end of the infusion, and 0.25, 0.5, 1, 2, 3, 4, 6, 8, 12 and 24 hours after the infusion. Protein-free filtrates of sera collected 0.25, 1 and 3 hours after the end of the infusion were assayed for aztreonam and clindamycin. The urines collected during the -8 to 0, 0 to 2, 2 to 4, 4 to 8, 8 to 24 hour periods after infusion were assayed for aztreonam and clindamycin. All subjects received physical examinations, 12-lead electrocardiograms, and a battery of tests of blood and urine chemistry, hematology and serum enzymes reflecting liver function.

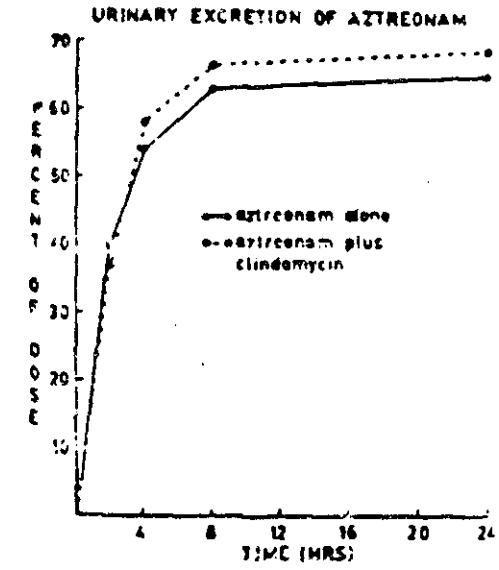
All pharmacokinetic parameters examined, except for urinary excretion, were not different, when comparisons were made between single drugs and the agents in combination. In the case of urinary excretion, very small, but statistically significant increases occurred for each drug, when the antibiotics were given in combination. These changes were clinically insignificant.

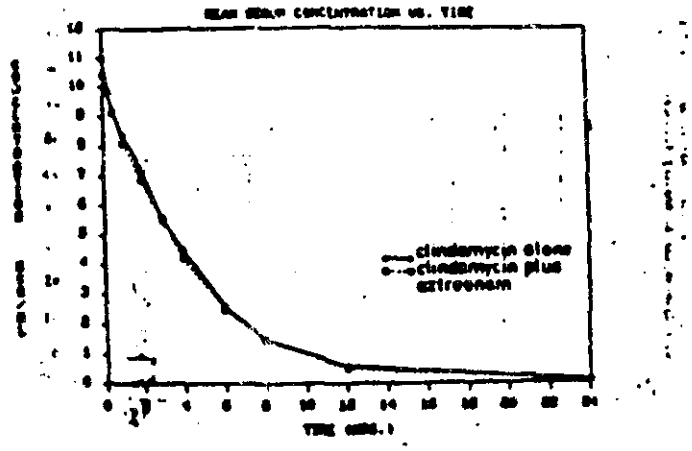
Apart from taste disturbance of 9 minutes to 2 hours duration in 6 subjects receiving combined therapy, which is probably just physical tasting of drugs circulating in the blood or excreted in the saliva, there were no untoward effects.

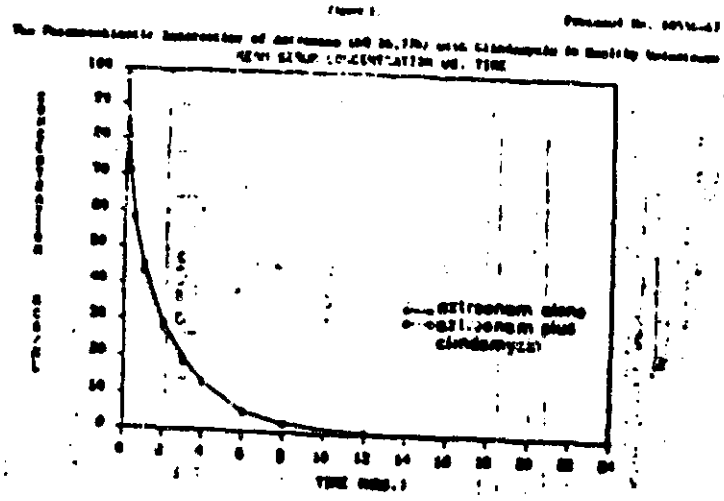
Pharmacokinetic parameters are summarized in the table below.

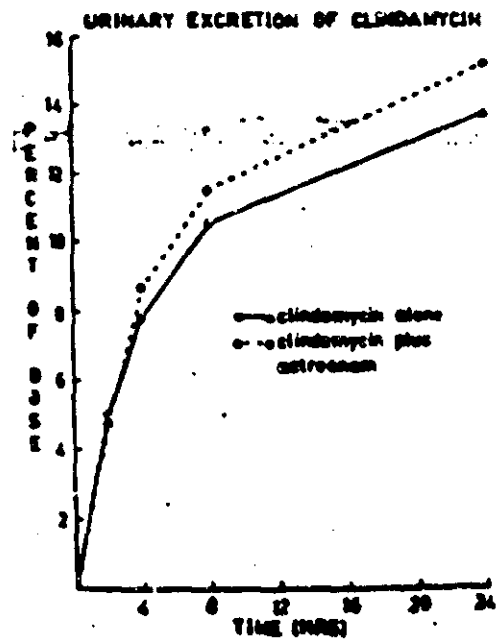
	Aztreonam Alone	Clindamycin Alone	Aztreonam plus Clindamycin: Aztreonam	Aztreonam plus Clindamycin: Clindamycin
Cmax (µg/ml)	90.5 ± 3.6	11.1 ± 0.5	96.2 ± 3.4	10.5 ± 0.8
AUC 0-24 hr (µg hr/ml)	170.2 ± 7.2	46.1 ± 5.6	172.5 ± 6.4	46.2 ± 6.8
t½ (hours)	1.66 ± 0.04	3.11 ± 0.38	1.68 ± 0.03	3.20 ± 0.52
Concentration in drug-containing effusate at 3 minutes (µg/ml)	29.91 ± 1.93	0.25 ± 0.04	30.0 ± 1.46	0.25 ± 0.04
Urinary Excretion 0-24 hr (percent of dose)	65.0 ± 2.0	12.7 ± 0.8	68.4 ± 2.0[a]	16.2 ± 0.8[a]

[a] Statistically different from corresponding mean for aztreonam or clindamycin alone, $p < 0.05$.









SECTION

Division of Medical Affairs

PRODUCT CODE
M69-860

PRODUCT OR NO. OR PROJECT CODE
SQ 26,776 (Aztreonam)

TITLE

Report on the Pharmacokinetic Interaction of Aztreonam (SQ 26,776) with Metronidazole in Healthy Volunteers

Study Protocol # 18,554-48

AUTHORS William A. Creasey, D.Phil., Michelle A. Stern, B.A. and Janice Lux, B.S., M.P.H.

INVESTIGATORS A. Arthur Sugerman, M.D., The Medical Center at Princeton, Princeton, N.J., Thomas Platt, Ph.D. and John Adamovics, Ph.D., The Squibb Institute for Medical Research, New Brunswick, N.J.

ABSTRACT

Page 19 to 30 yes 20

Nine (9) healthy male volunteers were enrolled in this single-dose, three-way, balanced crossover study. Each subject received, in randomized order, a single 30-minute intravenous infusion of aztreonam (1000 mg), metronidazole (500 mg), or aztreonam (1000 mg) plus metronidazole (500 mg) simultaneously, on each of 3 study days separated by a 7-day washout period. Concentrations of aztreonam and metronidazole were assayed in serum samples collected at the end of the infusion, and 0.25, 0.5, 1, 2, 3, 4, 6, 8, 12 and 24 hours after the infusion. Protein-free filtrates of sera collected 0.25, 1 and 3 hours after the end of the infusion were assayed for aztreonam. The urines collected during the -8 to 0, 0 to 2, 2 to 4, 4 to 8 and 8 to 24 hour periods after infusion were assayed for aztreonam and metronidazole. All subjects received physical examinations, 12-lead electrocardiograms, and a battery of tests of blood and urine chemistry, hematology and serum enzymes reflecting liver function.

Mean serum levels of aztreonam were slightly higher after administration of the monobactam alone than when it was given in combination with metronidazole, but this trend was statistically significant only immediately after the infusion. However, although this difference was statistically significant, it was of such a low order of magnitude as to be clinically irrelevant.

Serum levels of metronidazole were unaffected when the two antibiotics were given in combination. Values for the C_{max}, the areas under the curve and the serum half-lives showed no significant differences between monotherapy and the combination.

Urinary excretion of aztreonam reached 63.2 and 62.0 percent of administered dose after single drug or combined treatment, respectively. In the case of metronidazole, the corresponding figures were 22.4 and 21.6 percent, respectively. No adverse reactions were encountered. The table below summarizes the pharmacokinetic data.

	Aztreonam Alone	Metronidazole Alone	Aztreonam plus Metronidazole: Aztreonam	Aztreonam plus Metronidazole: Metronidazole
C_{max} (µg/ml)	97.9 ± 4.2	5.7 ± 0.28	98.3 ± 2.9*	6.0 ± 0.43
Serum-protein Binding (percent at 0.25 hr)	61.5 ± 1.2	N.D.	61.2 ± 1.6	N.D.
AUC 0-24 (µg.hr/ml)	161.9 ± 8.4	36.6 ± 3.1	163.9 ± 8.5	36.3 ± 1.6
$t_{\frac{1}{2}}$ (hours)	1.6 ± 0.07	10.8 ± 0.6	1.7 ± 0.04	12.2 ± 0.6
Urinary Excretion 0-24 hr (percent of dose)	63.2 ± 2.8	22.4 ± 0.7	62.0 ± 1.1	21.6 ± 1.6

*Statistically significantly less than mean for aztreonam alone ($p < 0.05$).

N.D. not determined.

FIGURE 1

THE PHARMACOKINETIC INTERACTION OF AZTREONAM WITH METRONIDAZOLE IN HEALTHY VOLUNTEERS

SERUM CONCENTRATIONS OF AZTREONAM

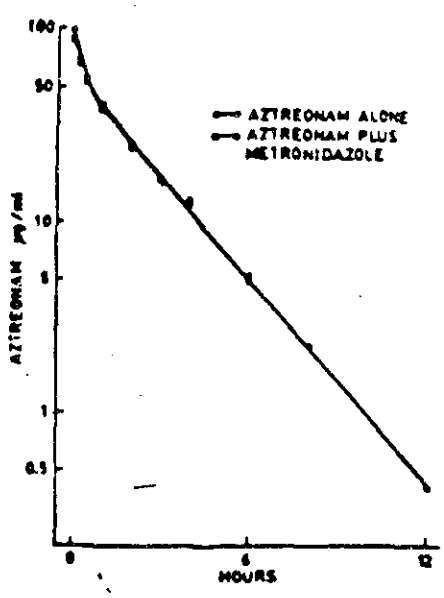


FIGURE 2

THE PHARMACOKINETIC INTERACTION OF AZTREONAM WITH METRONIDAZOLE IN HEALTHY VOLUNTEERS

CUMULATIVE URINARY EXCRETION OF AZTREONAM

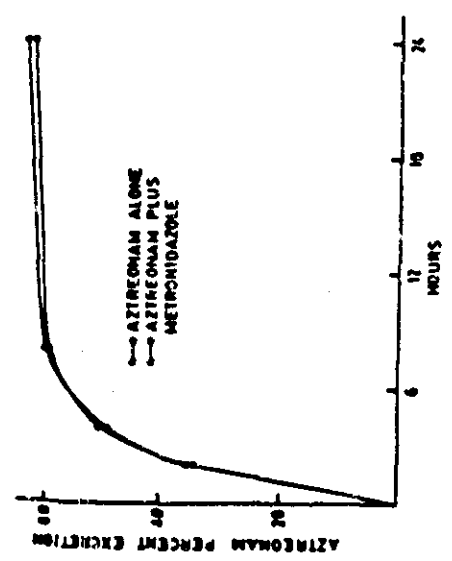


FIGURE 3

THE PHARMACOKINETIC INTERACTION OF AZTREONAM WITH METRONIDAZOLE IN HEALTHY VOLUNTEERS

SERUM CONCENTRATIONS OF METRONIDAZOLE

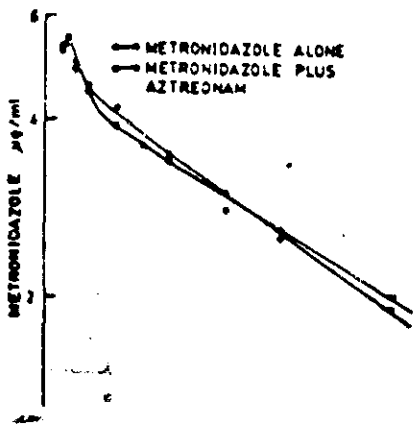
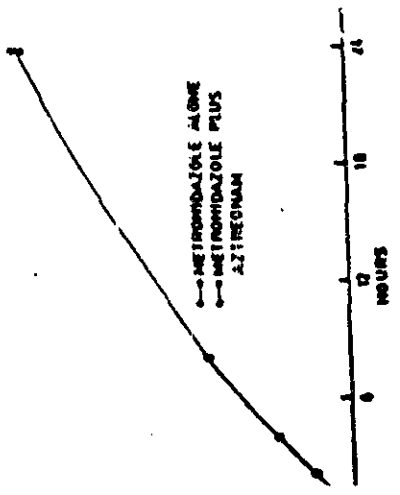


FIGURE 4

THE PHARMACOKINETIC INTERACTION OF AZTREONAM WITH METRONIDAZOLE IN HEALTHY VOLUNTEERS

CUMULATIVE URINARY EXCRETION OF METRONIDAZOLE



Department of Clinical Pharmacology — Date: [illegible], 1985

SECTION: Division of Medical Affairs

PROJECT CODE: MAB-860

PROJECT, SQ NO OR PROJECT NAME: SQ 26,776 (Aztreonam)

TITLE

Report on the Pharmacokinetic Interaction of Aztreonam (SQ 26,776) with Nafcillin in Healthy Volunteers

Study Protocol # 18 554-49

AUTHORS: William A. Creasey, M. Ed., Michelle A. Stern, B.A. and Janice Lux, B.S., M.P.H.

INVESTIGATORS: A. Arthur Sugerman, M.D., The Medical Center at Princeton, Princeton, New Jersey 08540; John Adamovics, Ph.D., and Thomas R. Platt, Ph.D., Squibb Institute for Medical Research, New Brunswick, New Jersey 08903

ABSTRACT

Nine (9) healthy male volunteers were enrolled in this single-dose, three-way, balanced crossover study. Each subject received, in randomized order, a single 30-minute intravenous infusion of aztreonam (1000 mg), nafcillin (500 mg) or aztreonam (1000 mg) plus nafcillin (500 mg) simultaneously, on each of 3 study days separated by a 7-day washout period. Concentrations of aztreonam and nafcillin were assayed in serum samples collected at the end of the infusion, and 0.25, 0.5, 1, 2, 3, 4, 6, 8, 12 and 24 hours after the infusion. Protein-free filtrates of sera collected 0.25, 1 and 3 hours after the end of the infusion were assayed for aztreonam and nafcillin. The urines collected during the -8 to 0, 0 to 2, 2 to 4, 4 to 8 and 8 to 24 hour periods after infusion were assayed for aztreonam and nafcillin. All subjects received physical examinations, 12-lead electrocardiograms, and a battery of tests of blood and urine chemistry, hematology and serum enzymes reflecting liver function.

All pharmacokinetic parameters examined, with the sole exception of aztreonam levels in the 0.25 hour protein-free filtrate, were not significantly different, when comparisons were made between single drugs and the agents in combination. In the case of the protein-free filtrate, there was a 15 percent increase in the aztreonam level and a small decrease in the percent bound to protein when the drugs were co-administered.

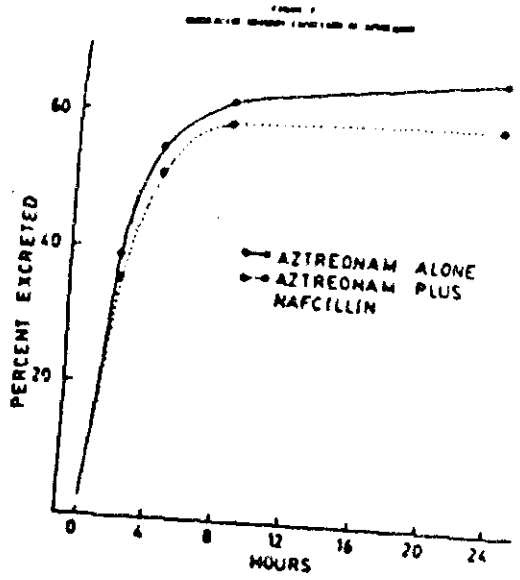
Apart from a taste disturbance of 2 minutes duration in one subject, which was probably just physical tasting of drug circulating in the blood or excreted in the saliva, there were no untoward effects.

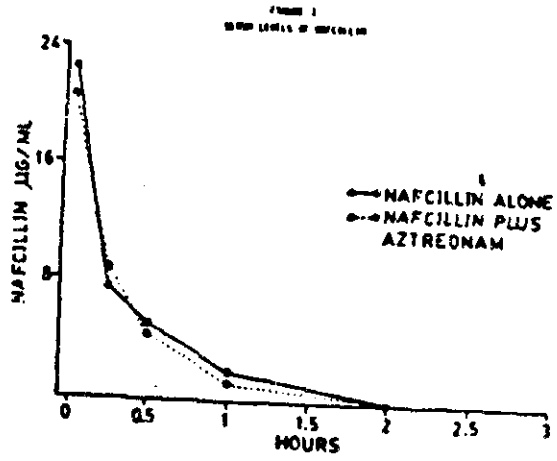
Pharmacokinetic parameters* are summarized in the table below.

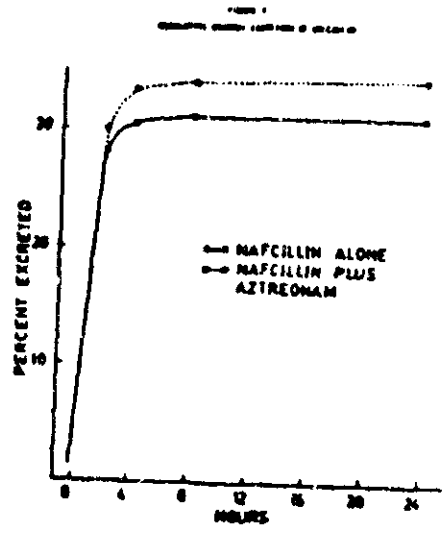
	Aztreonam Alone	Nafcillin Alone	Aztreonam plus Nafcillin	
			Aztreonam	Nafcillin
C_{max} (ug/ml)	103.9 ± 7.8	22.5 ± 2.2	97.4 ± 5.3	20.6 ± 3.6
AUC 0-24 hr (ug.hr/ml)	170.3 ± 12.1	8.2 ± 0.9	168.2 ± 13.9	7.3 ± 1.1
$t_{\frac{1}{2}}$ (hours)	1.60 ± 0.08	0.38 ± 0.05	1.60 ± 0.09	0.38 ± 0.02
Protein-free filtrate (ug/ml at 0.25 hr)	25.9 ± 1.9[a]	0.7 ± 0.1	29.7 ± 1.4	1.0 ± [illegible]
Serum-protein Binding (percent at 0.25 hr)	63.8 ± 1.6[a]	86.2 ± 4.9	69.1 ± 1.0	87.2 ± [illegible]
Urinary Excretion 0-24 hr (percent of dose)	67.1 ± 3.9	31.3 ± 6.4	60.4 ± 3.7	34.8 ± 4.6

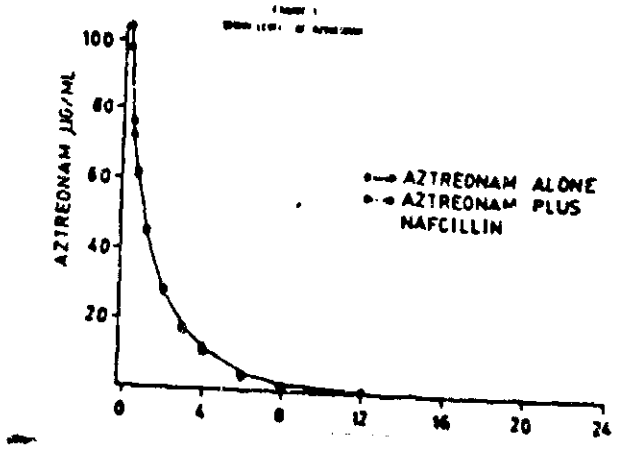
[a] Different from value for combined treatment at $p < 0.05$

*In one subject, serum level data after infusion of either antibiotic alone were uncharacteristic of intravenous drug administration. These data were, therefore, excluded from statistical analyses and the above-listed kinetic parameters are based on eight subjects.









SECTION

Division of Medical Affairs

MNB-860

SQ 26,776 (Aztreonam)

TITLE

Report on the Pharmacokinetic Interaction of Aztreonam (SQ 26,776) with Cephradine in Healthy Volunteers

Study Protocol # 18,554-59

AUTHORS: William A. Creasey, D.Phil., Michelle A. Stern, B.A. and Janice Lue, B.S., M.P.H.

INVESTIGATORS: A. A. Sugerman, M.D., The Medical Center at Princeton, Princeton, N.J., 08540, T. B. Platt, Ph.D., K. Dhruv, Ph.D., J.A. Manning, and J. Adamovics Ph.D., Squibb Institute for Medical Research, New Brunswick, N.J. 08903

ABSTRACT

→ ages 19 to 34 years

Nine (9) healthy male volunteers completed this single-dose, three-way, balanced, crossover study. Two additional subjects enrolled initially failed to complete the study. Each subject received, in randomized order, a single 30-minute intravenous infusion of aztreonam (1000 mg), cephradine (1000 mg) or aztreonam (1000 mg) plus cephradine (1000 mg) simultaneously, on each of 3 study days separated by a 7-day washout period. Concentrations of aztreonam and cephradine were assayed in serum samples collected at the end of the infusion, and 0.25, 0.5, 1, 2, 3, 4, 6, 8, 10 and 12 hours after the infusion. Protein-free filtrates of sera collected 0.25, 1 and 3 hours after the end of the infusion were assayed for aztreonam. The urines collected during the -8 to 0, 0 to 2, 2 to 4, 4 to 8, and 8 to 24 hour periods after infusion were assayed for aztreonam and cephradine. All subjects received physical examinations, 12-lead electrocardiograms, and a battery of tests of blood and urine chemistry, hematology and serum enzymes reflecting liver function.

All pharmacokinetic parameters examined, except for serum binding of aztreonam, were not different, when comparisons were made between single drugs and the agents in combination. In the case of serum binding, very small, but statistically significant increases occurred for aztreonam when the antibiotics were given in combination. These changes were clinically insignificant.

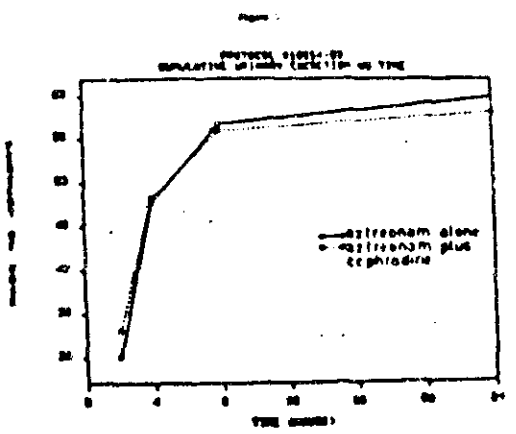
No drug-related adverse effects were noted.

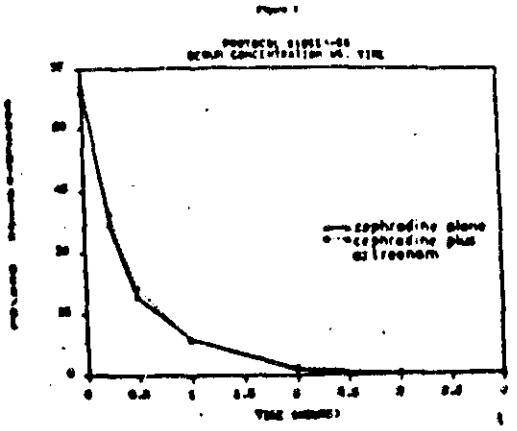
Pharmacokinetic parameters are summarized in the table below.

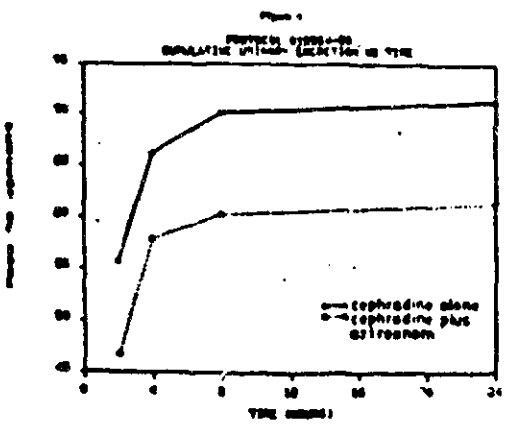
	Aztreonam Alone	Cephradine Alone	Aztreonam plus Cephradine: Aztreonam	Aztreonam plus Cephradine: Cephradine
Cmax (µg/ml)	97.4 ± 4.0	70.5 ± 2.5	95.7 ± 5.7	68.9 ± 6.0
AUC 0-24 hr (µg.hr/ml)	171.3 ± 5.4	33.5 ± 1.9	163.6 ± 6.6	35.5 ± 4.3
t½ (hours)	1.71 ± 0.07	0.40 ± 0.03	1.61 ± 0.08	0.42 ± 0.04
Concentration in Protein-free filtrate at 15 minutes (µg/ml)	29.33 ± 0.33	N.D.	28.48 ± 1.03	N.D.
Serum-protein Binding (percent at 15-minutes)	60.6 ± 1.1	N.D.	59.8 ± 1.5	N.D.
Serum-protein Binding (percent at 3 hours)	61.0 ± 0.8	N.D.	64.1 ± 0.7[a]	N.D.
Urinary Excretion 0-24 hr (percent of dose)	82.7 ± 5.6	71.3 ± 2.8	80.9 ± 4.7	61.4 ± 6.7

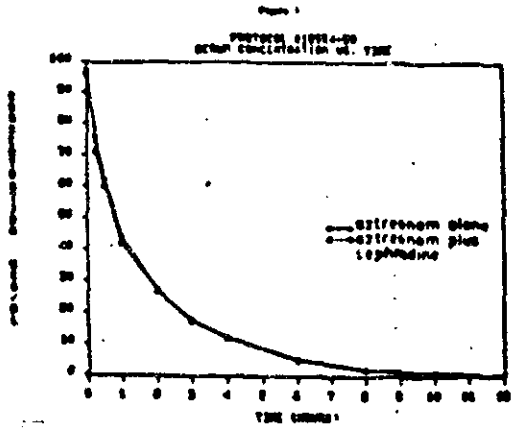
[a]Statistically different from corresponding mean for aztreonam alone, $p < 0.05$.

ND - not determined.









DEPARTMENT
Department of Clinical Pharmacology

SECTION
Division of Medical Affairs

DATE OR PERIOD COVERED
July 15, 1983

PROJECT CODE
MLB-800

PRODUCT, SQ NO OR PROJECT NAME
SQ 26,776 (Aztreonam)

TITLE:
Report on Single Intravenous Dose Safety and Pharmacokinetic Study of Aztreonam (SQ 26,776) in Patients with Normal or Inflamed Meninges

Study Protocol # 18,554-29

AUTHOR(S) Edward A. Swabb, M.D., Ph.D., and May Frantz, Ph.D.

INVESTIGATORS Richard J. Duma, M.D., Ph.D., Medical College of Virginia, and Thomas B. Platt, Ph.D., The Squibb Institute.

ABSTRACT

Aztreonam was administered as a single, 5-minute intravenous infusion of 2000 mg to 30 patients (25 evaluable) with normal meninges and 10 patients (9 evaluable) with meningeal inflammation to investigate penetration of the monobactam into cerebrospinal fluid (CSF). To assess the safety of aztreonam, clinical laboratory tests were conducted before and after drug administration.

Aztreonam was well-tolerated by all patients and no adverse reactions were apparent.

The pharmacokinetic profile of aztreonam was assessed by measuring antibiotic concentrations in serum at 0.5 hours after drug administration, in CSF at a given time point (1 to 9 hours) after drug administration, and in serum at the time of spinal tap. Specimens were assayed for aztreonam using a microbiological assay, and for aztreonam and SQ 26,992 using a high-pressure liquid chromatography (HPLC) assay. Both methods gave consistent results for aztreonam concentrations, indicating the lack of detectable microbiologically active metabolites in the serum and CSF of patients with normal or inflamed meninges. The bioassay data were chosen for pharmacokinetic analysis, because the assay had a lower quantitation limit compared to the HPLC method.

The mean serum and CSF concentrations of aztreonam at various time intervals are given in Table I. The microbiologically inactive metabolite, SQ 26,992, resulting from the hydrolytic opening of the beta-lactam ring of aztreonam, was present at detectable levels (0.5 to 1 µg/ml) in the CSF of less than half of the patients studied.

Based on areas under the serum and CSF concentration-time curves, the CSF penetration of aztreonam in the absence of meningeal inflammation was 1.5% between 0.5 and 8 hours after dosing. Mean CSF levels of aztreonam in the absence of meningeal inflammation were 0.5 and 1 µg/ml at 1 and 4 hours, respectively, and in the presence of meningitis were 2 and 3.2 µg/ml at 1 and 4 hours, respectively, indicating that meningeal inflammation produced approximately 3 to 4 times higher CSF levels of aztreonam than values in the absence of inflammation.

The pharmacokinetics of aztreonam given as a single, 5-minute intravenous infusion indicated that a 2000-mg dose in adults would produce CSF concentrations of aztreonam that are potentially therapeutic for members of *Enterobacteriaceae* commonly responsible for gram-negative bacillary meningitis. The data from this study support the clinical investigation of aztreonam in the therapy of gram-negative meningitis, according to a 2-gram, q6h or q8h, intravenous dosing regimen in adult patients with normal renal function.

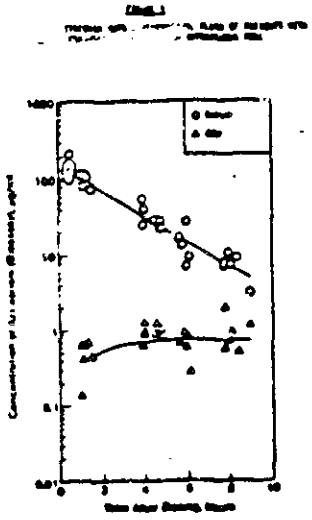
age 21 to 75 years

age 21 to 64 years,

Exclusion criteria included abnormal hepatic and renal function,

Patients with meningeal inflammation were given aztreonam in addition to other prescribed antibiotics as required.

Sterile powder blend of aztreonam and L-arginine (1.0/0.78).



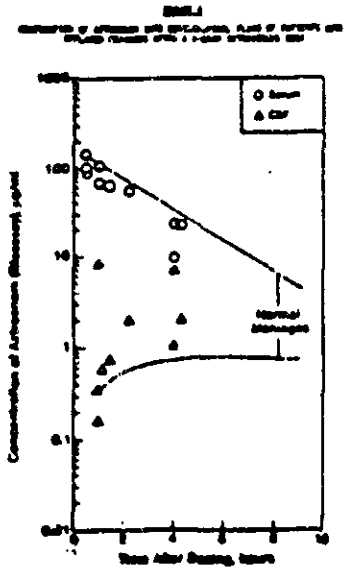


TABLE 1[a]

Number of Patients	0.5-Hr Serum Conc. µg/ml	Time of Tap, hr	Serum Conc. at Time of Tap, µg/ml	CSF Conc. µg/ml
Normal Meninges				
6	145±16 (121-169)	1.18±0.19 (1.00-1.47)	97.7±18.2 (73.2-119)	0.50±0.20 (0.14-0.69)
5	140±43 (109-216)	4.09±0.25 (3.93-4.53)	35.3±12.9 (25.1-55.4)	0.94±0.23 (0.63-1.28)
3	150±20 (128-167)	4.75±0.11 (4.63±4.83)	26.9±4.0 (22.2-29.4)	1.03±0.20 (0.83-1.22)
5	137±22 (100-157)	5.92±0.17 (5.67-6.10)	14.9±6.1 (6.99-27.6)	0.67±0.26 (0.28-0.93)
5	125±15 (105-138)	8.03±0.22 (7.82-8.37)	8.46±1.32 (7.40-10.50)	0.94±0.60 (0.51-1.97)
1	130	9.00	3.24	1.19
Inflamed Meninges				
5	126±18 (100-141)	1.09±0.18 (0.93-1.35)	88.4±21.5 (62.6-107)	1.98±3.44 (0.16-6.11)
1	139	2.17	54.7	1.96
3	112±27 (84.2-139)	4.15±0.16 (4.03-4.33)	18.0±7.2 (9.72-22.6)	3.22±2.99 (1.01-6.63)

[a]Values are mean ± SD (range). concentrations were determined by

Department of [illegible]

SECTION
Division of Medical Affairs

PROJECT CODE
MNB-860

PRODUCT OR GENERIC NAME
SQ 26,776 (Aztreonam)

TITLE

Report on a Single Intravenous Dose Safety and Pharmacokinetic Study of Aztreonam (SQ 26,776) in Patients with Inflamed Meninges

Study Protocol # 16,554-51

AUTHORS
Lawrence Y. Friedhoff, M.D., Ph.D., and Janice Lux, B.S., M.P.H.

INVESTIGATORS Richard L. Greenman, M.D., University of Miami, School of Medicine, Leon D. Sabath, M.D., University of Minnesota School of Medicine, and Thomas B. Platt, Ph.D., The Squibb Institute

ABSTRACT

Eleven patients aged 19 to 71 years with meningeal inflammation were given a single 5-minute, 2-gram intravenous infusion of aztreonam. Serum and cerebrospinal fluid (CSF) were assayed for aztreonam content by microbiological and high pressure liquid chromatographic (HPLC) assay. The results of the two assay methods were in good agreement. The HPLC assay results are discussed below. where gaps are required, other authors

Aztreonam was generally well tolerated. One patient had two minor adverse reactions. The first consisted of a "bad taste in the mouth", which began immediately after drug infusion, lasted 5 minutes and resolved without therapy. The second consisted of a feeling of "floating and loss of contact with concrete things." This sensation began immediately after dosing, lasted 30 minutes, resolved without therapy and occurred shortly after the patient had received a large dose of intravenous penicillin.

Four patients were inadvertently enrolled in the study who did not meet the entry criteria because of elevated SGOT or SGPT, elevated CSF red cell count, or cardiovascular instability. These patients were classified as "excluded". Since the serum and CSF aztreonam concentrations found in these four patients were similar to those found in the other patients, the results for included and excluded patients were pooled. Concentrations of aztreonam in serum and CSF are summarized in Table I.

Serum concentrations of aztreonam averaged 101 mcg/ml, 41 mcg/ml and 15 mcg/ml at 0.5, 2 and 4 hours after dosing, respectively). Aztreonam was detectable in the CSF at 0.5 hours after dosing. The mean CSF concentration averaged 1.36 mcg/ml, 2.79 mcg/ml, 4.60 mcg/ml and 3.31 mcg/ml at 1, 2, 4 and 8 hours after administration (see Table I). Maximal concentrations occurred at 2 to 4 hours after dosing. In two patients (nos. 009 and 012), multiple CSF samples were obtained from CSF drains. The data from these two patients confirm the fact that the maximal CSF concentration was achieved at 2 to 4 hours after dosing.

SQ 26,992 (the open beta-lactam ring hydrolysis product of aztreonam) was assayed by HPLC in both serum and CSF. In general, the concentration was below 1 mcg/ml, the limit of quantification of the assay.

The results of this study demonstrate that a single intravenous 2-gram dose of aztreonam generally produces CSF concentrations two to five times the MIC_{90} for common *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Proteus vulgaris*, *Proteus rettgeri*, *Providencia stuartii*, *Salmonella sp.*, *Hemophilus influenzae* and *Neisseria gonorrhoeae*. These concentrations are maintained for up to 8 hours.

These results support clinical investigation of a q6h to q8h 2-gram intravenous dosage regimen in patients with normal renal function and gram-negative meningitis due to susceptible organisms.

TABLE I

Protocol No. 16554-51 (Revised 6/22/83)

HPLC AZTREONAM ASSAY RESULTS (mcg/ml)

Patient Number	SERUM TIME (hours) 0	0.5	1	2	4	6	8	CSF TIME (hours) 0.5	1	2	4	6	8
006 I	-	120		22.7									
007 I	0	128											
008 E	0	117		34.9									
009 I	0	84.1		38.0							16.1		
010 I	0	66.4		27.7	21.2					0.67			
011 I	0	66.4		34.9	9.66					1.77			
013 I	0	124		37.8								5.27	
017 I	0	136		38.9								2.36	
MEAN (I)	0	108		44.1	17.8	-	-	-	-	2.78			
										2.84			
004 E	0	73.7	129	38.4	5.95	0	0	0.61	1.35	3.33	2.51	1.06	
010 E	0	86.9		29.9									
012 E	0				27.0					0.76[a]			
[b]012 E	279	74.3	59.8	32.6	31.0	0	0					7.83	
MEAN (E)	-	78.6	-	33.2	-	-	-	-	1.35	1.66	1.76	1.05	
										2.14			
OVERALL MEAN		101	-	41.4	14.8				1.36	1.92	2.14	3.31	
									1.36	2.79	4.60	3.31	

I - Included; E - Excluded

a - Diagonal result - no HPLC assay done

b - Patient 012 had a positive pre-dose serum aztreonam concentration, probably due to mislabeling of sample tubes. Serum data for this patient is excluded from the MEAN.

Mean 4 and 8 hour CSF levels may be over exaggerated due to an outlier of 16.1 mcg/ml at 4 hours and 7.83 mcg/ml at 8 hours.

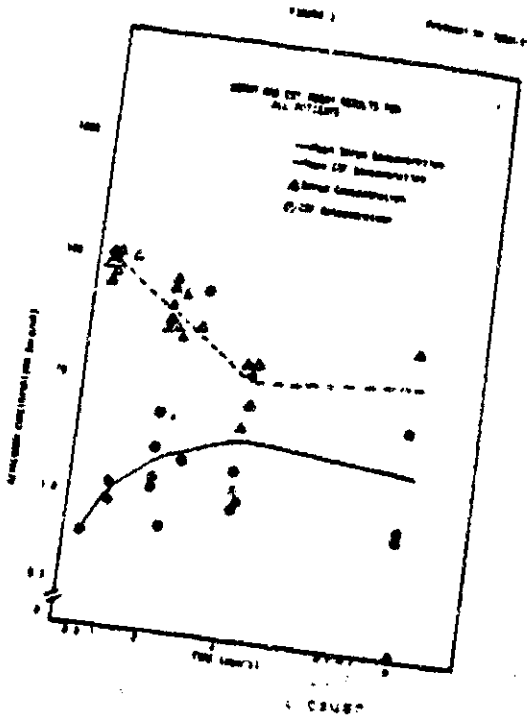
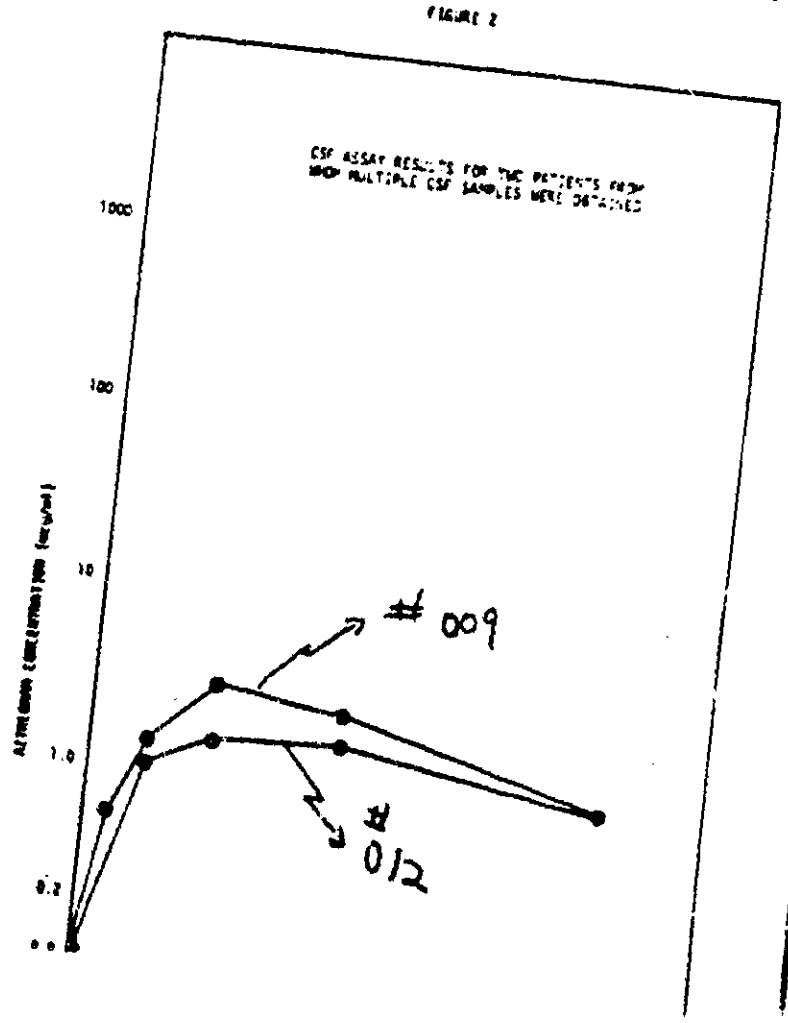


FIGURE 2



Department of Clinical Pharmacology

SECTION
Division of Medical Affairs

PROJECT CODE
MNB-840

PRODUCT, NAME OR PROJECT CODE
SQ 26,776 (Aztreonam)

TITLE

Report on Biliary Excretion of Aztreonam in Patients with T-tubes Inserted in the Common Bile Duct Study Protocol # 18,554-12

AUTHORS

Richard G. Devlin, Ph.D., Michelle A. Stern, B.A. and Janice Lux, B.S.N.; M.P.H

INVESTIGATORS

J. Levi, M.D. and O. Martinez, Ph.D., University of Miami, Miami, Fla. and T. Platz, Ph.D., Squibb Institute for Medical Research, New Brunswick, N.J.

ABSTRACT

Aztreonam was administered as a single, 1 gm, 2-minute intravenous injection to 14 volunteers. Two groups of volunteers were studied. Group A consisted of 10 post-cholecystectomy patients, each of whom had a T-tube in place for partial collection of bile output. Group B consisted of 4 presurgery patients, each of whom had carcinomas of the pancreas or ampulla. Patients in Group B also had a T-tube in place but, unlike patients in Group A, complete, quantitative collections of total bile output were accomplished in Group B patients.

Samples of serum, urine and bile were collected from each patient at various times up to 12 hr after injection of aztreonam. For patients in both groups, small aliquots of bile were obtained at precise times after aztreonam injection. Patients in Group B also had quantitative collections of total bile output, at various time intervals, up to 12 hr after aztreonam administration.

Pharmacokinetic analysis of aztreonam in bile for both groups of patients is shown in Table 1 (microbiological assay).

Age 25 to 75 years.

TABLE 1

Pharmacokinetic Analysis of Aztreonam in Human Bile

Parameter	Group A[a]	Group B[a]
AUC_{0-12hr}, ugxhr/ml	176.9±32.2	36.6*±9.9
Cmax, ug/ml	42.9±7.9	13.5**±4.2
Tmax, hr	2.4±0.2	1.0**±0.4
t½β, hr	2.3±0.3	3.0±0.1
12-hr biliary excretion % of dose	-	0.18±0.06

a) Patients in Group A (N=10) were post-cholecystectomy; patients in Group B (N=4) were awaiting surgery; both groups had T-tubes in place during study but only in Group B patients was quantitative bile collection possible.

*significantly different from mean for group A, $p < 0.05$

**significantly different from mean for group A, $p < 0.01$

Much less aztreonam was excreted in the bile by patients in Group B as compared to those in Group A. Patients in Group B had total obstruction of biliary flow before placement of the T-tube (the study was done 24 hr after T-tube placement). Biliary obstruction is known to inhibit antibiotic excretion in the bile. Thus, the lower levels of biliary aztreonam excretion in Group B may suggest that the liver in those patients had not fully recovered its excretory function at 24-36 hours after decompression by the T-tube.

Cumulative bile collections in Group B patients allowed a determination of total biliary excretion of aztreonam in those patients. A mean (±SEM) value of 0.18 ± 0.06 percent of the administered aztreonam dose was excreted in the bile of Group B patients.

HPLC analysis of bile samples revealed no quantitatively detectable SQ 26,992 in the bile of 13 patients in this study. One patient (14) had small amounts of SQ 26,992 in the bile, ranging from 2.2 to 6.0 ug/ml. This patient was the only one in the study diagnosed as having adenocarcinoma of the ampulla.

These data may be compared with those from a previously documented study in which it was found that in healthy subjects about 1.5 and 3.5% of an i.v. dose was excreted in the feces as aztreonam and SQ 26,992, respectively.

Aztreonam was well tolerated by volunteers in this study; patient 6 in Group A experienced mild nausea approximately 2 hours after the administration of aztreonam, which lasted less than 24 hours. Aztreonam was considered by the investigator to be a possible cause of the nausea.

Mean (±SEM) Biliary Concentrations (ug/ml) of Aztreonam

Time After Injection (hr)	Group A (N=10)	Group B (N=4)
0.5	8.8[a]±4.7	10.0±5.7
1.0	27.6±7.1	10.8±3.0
2.0	39.3±6.4	7.3[c]±1.7
3.0	29.9±6.2	-
4.0	20.1±6.3	6.8±1.1
6.0	14.5±3.5	2.3±0.9
8.0	5.8±1.5	1.1±0.6
12.0	3.0±0.8	0.2±0.2

a) Includes estimated concentration of 0.2 ug/ml for Patient No. 9.

b) Includes estimated concentration of 0.8 ug/ml for Patient No. 3.

c) Includes estimated concentration of 7.4 ug/ml for Patient No. 13.

TABLE 33

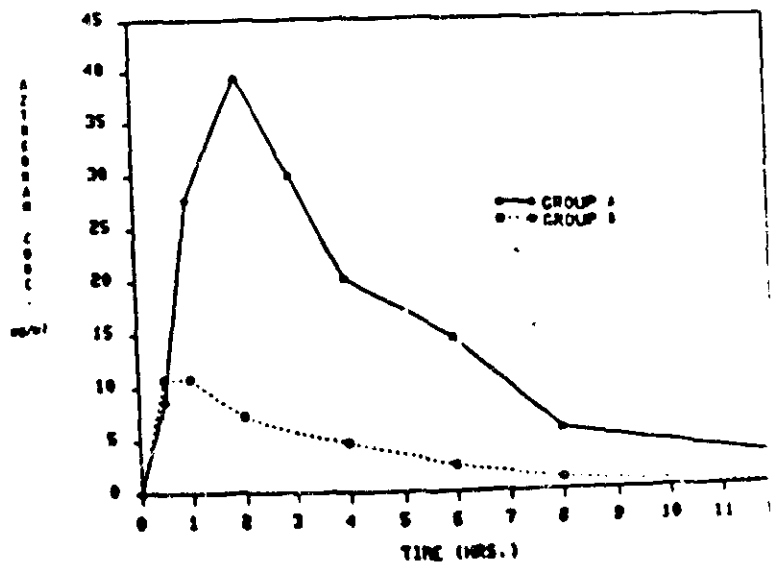
Maximum Biliary Concentration (ug/ml) of Aztreonam

Patient No.	Group A Cmax	Patient No.	Group B Cmax
1		11	
2		12	
3		13	
4		14	
5			
6			
7			
8			
9			
10			
Mean (±SEM)	42.9±7.9		13.5*±4.2

*significantly different from mean for Group A, $p < 0.05$

Fig. 1.

BILIARY EXCRETION OF AZTREONAM, MEAN CONCENTRATIONS VS TIME



SECTION

Division of Medical Affairs

MNB-860

SQ 26,776 (Aztreonam)

TITLE

Report on a Single Intravenous Dose Safety and Pharmacokinetic Study of Aztreonam (SQ 26,776) Penetration Into Human Bronchial Secretion

Study Protocol #18,554-54

AUTHORS:

Lawrence T. Friedhoff, M.D., Ph.D., and Cecelia Vertucci, B.A.

INVESTIGATORS:

Douglas L. Bechard, M.D. and Stephen S. Hawkins, M.D., Erlanger Medical Center, Chattanooga, Tennessee, and T.B. Platt, Ph.D., The Squibb Institute

ABSTRACT

ABSTRACT

Ages 20 to 65 years

Ten intubated patients were each given a single 2-gram intravenous infusion of aztreonam over 5 minutes. Serum and bronchial secretion samples were obtained after dosing. Aztreonam was well tolerated by all patients, and no adverse reactions were noted.

The results of serum and bronchial secretion assays are shown in Table 1 (next page). Some or all assay data obtained from each of 4 patients were excluded because of the presence of aztreonam in the "pre-dose" bronchial secretion sample, blood in a bronchial secretion sample, or because the patient did not meet study entry criteria. Samples could not always be obtained precisely at the scheduled times due to clinical considerations. Actual sampling times were rounded to the nearest scheduled time for the purpose of Table 1. The average serum concentrations for the included patients were 83.4, 43.0, 25.4 and 8.9 mcg/ml at 0.5, 2.0, 4.0 and 8.0 hours after dosing, respectively. The average bronchial secretion concentrations for the included patients were 3.3, 4.8, and 1.9 mcg/ml at 2, 4 and 8 hours after dosing, respectively. Thus, the maximal average bronchial secretion concentration was achieved at approximately 4 hours after dosing. All patients (included and excluded) had at least one bronchial secretion sample with a concentration equal to or above 2.7 mcg/ml at some time during the study (2, 4 or 8 hours after dosing).

The mean of the ratios of the bronchial secretion to serum aztreonam concentrations was 0.25 at 4 hours after dosing, when the mean bronchial secretion concentration was maximal.

The MIC_{90} of aztreonam for commonly encountered *Enterobacteriaceae* is reported to be less than or equal to 1.0 mcg/ml. Thus, the results of this study support clinical investigation of a q6-q8h, 2-gram intravenous dosage regimen for patients with normal renal function and serious pneumonia due to susceptible organisms.

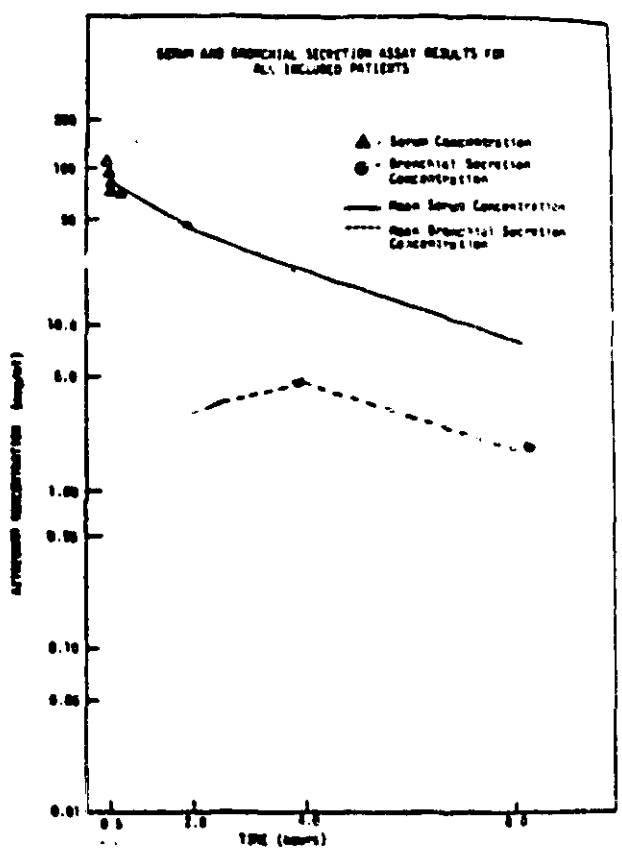
TABLE 1

SERUM AND BRONCHIAL SECRETION BIOASSAY RESULTS

PATIENT NUMBER	BRONCHIAL SECRETION PURULENCE	SERUM TIME (HOURS) 0	0.5	1.0	2	4	8	BRONCHIAL SECRETION TIME (HRS) 0	2	4	8
001 E	•										
002 E	N.D.										
003 I	N.D.										
004 I	•										
005 I	•										
006 I	•										
007 I	•										
008 I	•										
009 E	•										
010 I[a]	•										
RESULTS INCLUDED PATIENTS		0	83.4	-	43.0	25.4	8.9	0	3.3	4.8	1.9

I - Included patient
E - Excluded patient
• - Purulent bronchial secretion
N.D. - Sample not obtained
[a] - Eight hour bronchial secretion data excluded
[b] - The presence of aztreonam in the "predose" sample was confirmed by HPLC. This sample was probably inadvertently collected shortly after aztreonam administration.

FIGURE 1



SECTION

Division of Medical Affairs

MNB-860

PRODUCT, ITS RX, OR PRODUCT NAME

SQ 26,776 (Aztreonam)

TITLE

Report on Aztreonam Excretion in Human Milk

Study Protocol #18554-33

AUTHORS:

Richard G. Davis, Jr., Ph.D., May Frantz, Ph.D., and Michelle Stern, B.A.

INVESTIGATORS:

Paul M. Preiss, M.D., Los Angeles, California; T.B. Platt, Ph.D., The Squibb Institute

Ages 23 - 35 years

ABSTRACT

In this study, each of 12 normal, lactating subjects received a single 1-gm dose of aztreonam. Six (6) subjects received an intramuscular (i.m.) injection and 6 an intravenous (i.v.) injection. Sequential and concomitant serum and milk samples were obtained over the 8-hour period following the injection.

Mean bioavailability parameters for aztreonam in serum and milk are summarized below:

Parameter	Units	Intramuscular Injection Serum	Milk	Ratio[a] Milk/ Serum	Intravenous Injection Serum	Milk	Ratio Milk/ Serum
AUC_{0-8hr}	ug x hr/ml	173.0	1.5	0.009	182.6	1.0	0.005
C_{max}	ug/ml	42.6	0.3	0.007	126.2	0.2	0.002
T_{max}	hr	1.3	6.0	6.0	0.25*	2.4	9.6

a) Values based on mean of individual ratios.
* Initial blood sample was collected 15 min. after aztreonam injection.

Milk levels of aztreonam were much lower than serum levels at every sampling time. The AUC_{0-8hr} values and the C_{max} values for aztreonam in milk were less than 1% of those in serum after both i.m. and i.v. injections. T_{max} values were 6 and 10 times longer in milk than in serum after i.m. and i.v. injections respectively.

Assuming a large milk production of 1 liter per day and taking the C_{max} of aztreonam in milk (0.3 ug/ml after i.m. injection) as a mean concentration, the amount of aztreonam in the daily maternal milk would be about 300 ug or 0.3 mg. *

The low levels of aztreonam found in milk in this study, along with the previously documented very poor oral absorption of aztreonam, suggest that systemic ill effects would be unlikely to occur in a breast-feeding infant whose mother received a therapeutic dose of aztreonam. The low levels of aztreonam found in breast milk also suggest that insufficient aztreonam would be ingested by the nursing infant to produce untoward effects on intestinal flora. *2

No adverse reactions occurred in this study.

Table 3

MEAN (± SEM) SERUM AND MILK CONCENTRATIONS (ug/ml) OF AZTREONAM

Time After Injection	Intramuscular Injection		Intravenous Injection	
	Serum	Milk	Serum	Milk
0.25	30.0 ± 5.6	0.00[a]	126.3 ± 17.1	0.0
0.5	33.9 ± 6.1	0.00	75.9 ± 6.5	0.0
1.0	3... ± 3.9	0.00	52.4 ± 4.4	0.07[a] ± 0.04
1.5	40.0 ± 1.6	0.02[a] ± 0.02	37.4 ± 2.8	0.11 ± 0.05
2.0	35.0 ± 1.8	0.14[a] ± 0.04	29.4 ± 2.9	0.18 ± 0.06
3.0	28.8 ± 1.3	0.41 ± 0.19	19.1 ± 1.9	0.16 ± 0.06
4.0	20.2 ± 2.1	0.32 ± 0.08	14.0 ± 1.8	0.22 ± 0.06
6.0	11.6 ± 1.8	0.34 ± 0.08	6.2 ± 1.0	0.14 ± 0.05
8.0	6.0 ± 0.7	0.38 ± 0.07	2.8 ± 0.5	0.12 ± 0.04

[a] Mean based on 5 subjects

FIGURE 1

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AND MILK OF LACTATING NORMAL SUBJECTS AFTER INTRAMUSCULAR ADMINISTRATION

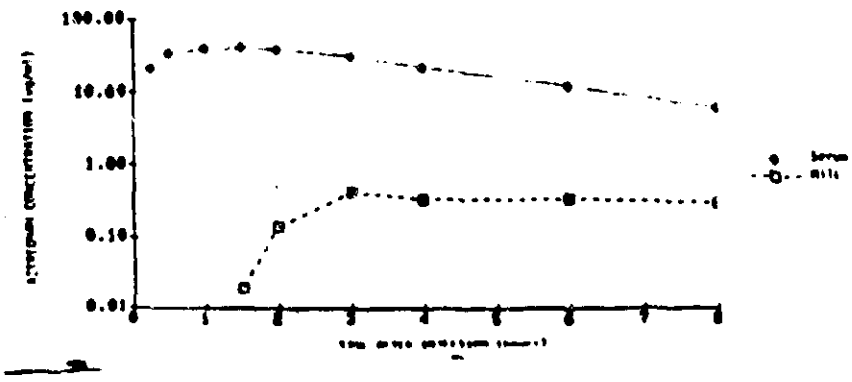
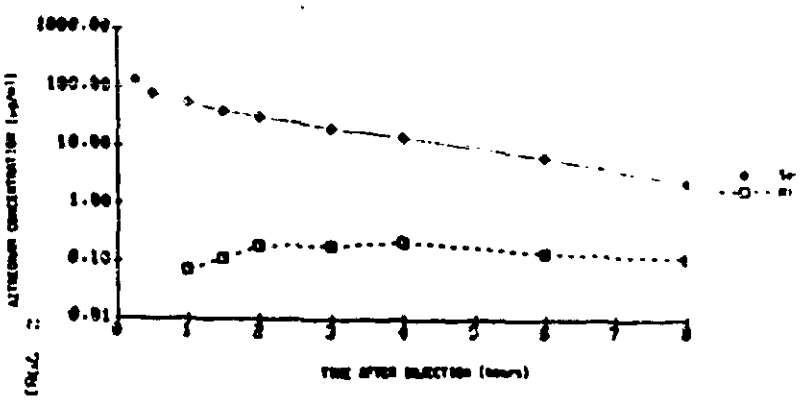


FIGURE 2

MEAN CONCENTRATIONS OF AZTREONAM IN SERUM AND MILK OF LACTATING NORMAL SUBJECTS AFTER INTRAVENOUS ADMINISTRATION



SECTION	PROJECT CODE
Division of Medical Affairs	MNB-860
	PRODUCT SQ NO. OR PROJECT LINE: SQ 26,776 (Aztreonam)

TITLE:

Report on the Determination of Aztreonam in Body Fluids in Mid-Term Pregnant Women.

Study Protocol #18,554-34

AUTHOR(S):
Richard G. Devlin, Jr., Ph.D., May Frantz, Ph.D., and Michelle Stern, B.S.

INVESTIGATOR(S):
Robert T. Hayashi, M.D., San Antonio, Texas; T.B. Platt, Ph.D., The Squibb Institute

ABSTRACT

In this study, aztreonam was administered as a single, 1-gm, intravenous injection to 12 mid-term pregnant subjects, who had elected to undergo therapeutic abortions. Labor was induced by injection of prostaglandin 2 hours after the aztreonam injection in 6 subjects (Group A) and 8 hours after the aztreonam injection in 6 other subjects (Group B). The abortion process took an average of 20.4 and 24.9 hours for subjects in Groups A and B respectively. Samples of maternal serum and amniotic fluid were collected before the abortion and samples of fetal serum and placenta were collected after fetal and placental expulsion.

Aztreonam levels in maternal serum were similar to those reported previously for normal subjects. Aztreonam was detected in amniotic fluid as early as 15 minutes after injection. The concentration of aztreonam in amniotic fluid increased over the entire collection period, reaching a mean (± SEM) concentration of 2.0 ± 0.4 ug/ml at 6 to 8 hours after injection for subjects in Group B. The latter value exceeded the mean (± SEM) aztreonam concentration in serum (0.9 ± 0.1 ug/ml) at 8 hours after injection.

The fetuses of 10 out of 12 subjects were exposed to aztreonam after intravenous injection in the mother. Mean (± SEM) aztreonam concentrations in fetal serum were 1.6 ± 0.4 and 0.5 ± 0.2 ug/ml for subjects in Groups A and B, respectively. The fetus of one subject in each group had no detectable aztreonam in the blood. Significantly more aztreonam was found in fetal serum samples derived from fetuses whose mothers were in Group A than those in Group B. This fact may be a reflection of the somewhat shorter mean (± SEM) time to fetal expulsion in Group A (20.4 ± 3.8 hr after aztreonam injection) than in Group B (24.9 ± 2.6 hr after injection).

Little or no aztreonam was found in the placentas of subjects in Group A (mean (± SEM) concentration was 0.1 ± 0.1 ug/gm) and no aztreonam whatever was found in the placentas of subjects in Group B.

Thus, aztreonam crossed the placenta and entered the fetal circulation after a single intravenous injection in the mother. In studies in laboratory animals given doses much higher than those used clinically, aztreonam was not fetotoxic or teratogenic. Nonetheless, fetal kidneys are immature and presumably would not clear aztreonam from the circulation as quickly as in the adult. Therefore caution should be exercised in the use of aztreonam in pregnant women.

Two subjects (Subjects 1 and 12) in this study had elevated levels of serum lactic dehydrogenase which the investigator thought were possibly related to aztreonam administration; however, no other significant changes in serum enzymes were observed. No other adverse effects of aztreonam were noted in this study.

TABLE 23

MEAN (± SEM) SERUM CONCENTRATIONS OF AZTREONAM (ug/ml)

Time After Injection (hr)	Group A[a]	Group B[b]
0.25	83.3 ± 5.2	77.4 ± 5.0
0.50	60.4 ± 3.3	58.4 ± 2.7
1.00	40.8 ± 2.6	36.9 ± 2.2
1.50	27.9 ± 1.6	26.2 ± 2.0
2.00	21.1 ± 1.1	18.8 ± 1.8
3.00	--	11.0 ± 1.2
4.00	--	6.4 ± 0.8
6.00	--	2.4 ± 0.3
8.00	--	0.9 ± 0.1

[a] Labor induced 2 hours after injection, serum collected only for 2 hours after injection.

[b] Labor induced 8 hours after injection, serum collected for 8 hours after injection.

TABLE 24

MEAN (± SEM) AMNIOTIC FLUID CONCENTRATIONS OF AZTREONAM (ug/ml)

Time After Injection (hr)	Group A[a]	Group B[b]
0.25	0.12 ± 0.04	0.19 ± 0.05
0.50	0.32 ± 0.07	0.32 ± 0.09
1.00	0.60 ± 0.10	0.64 ± 0.13
1.50	0.84 ± 0.16	0.85 ± 0.18
2.00	1.01 ± 0.16	1.13 ± 0.23
3.00	--	1.47 ± 0.25[c]
4.00	--	1.70 ± 0.32[c]
6.00	--	2.00 ± 0.36[c]
8.00	--	2.03 ± 0.35[c]

[a] Labor induced 2 hours after injection, amniotic fluid collected only for 2 hours after injection.

[b] Labor induced 8 hours after injection, amniotic fluid collected for 8 hours after injection.

[c] n=5 for these time points. Amniotic fluid samples from Subject 12 were thought to be contaminated with maternal blood and were not used in calculations of the mean.

TABLE 25

INDIVIDUAL AND MEAN (± SEM) CONCENTRATIONS OF AZTREONAM IN FETAL TISSUES

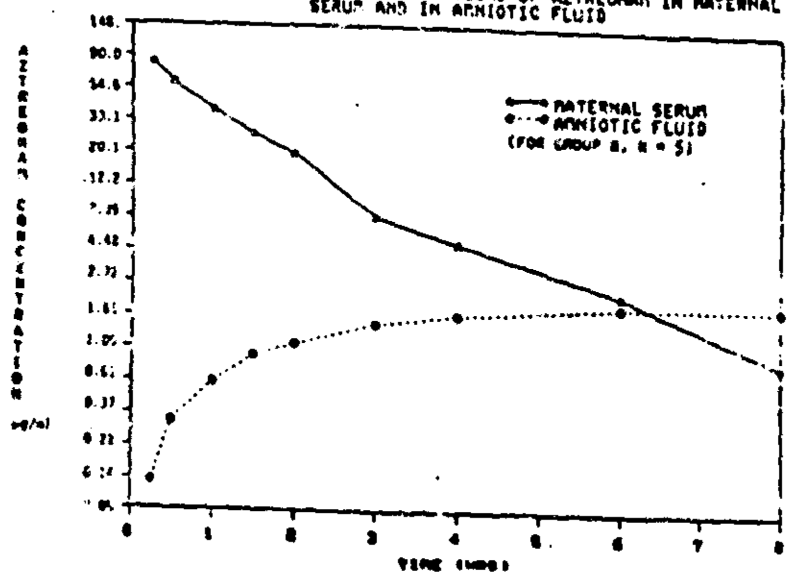
Subject No.[a]	Fetal Serum (ug/ml)	Placenta (ug/gm)
1		
2		
3		
4		
5		
6		
Mean (± SEM)	[b]1.6 ± 0.4	0.1 ± 0.1
7		
8		
9		
10		
11		
12		
Mean (± SEM)	0.5 ± 0.2	0.0 ± 0.0

[a] [illegible]

[b] [illegible]

FIGURE 3.

PROTOCOL #18554-34: CONCENTRATIONS OF AZTREONAM IN MATERNAL SERUM AND IN AMNIOTIC FLUID



SECTION:
Division of Medical Affairs

PROJECT CODE:
RNB-860

PRODUCT, ES NO., OR PROJECT NAME:
SQ 26,776 (Aztreonam)

TITLE:
Report on Intravenous Aztreonam Single Dose Pharmacokinetic and Tissue (Blister Fluid) Penetration Study.

Study Protocol # 18,554-39

AUTHOR(S):
Edward A. Swabb, Ph.D., M.D., and Cecelia Vertucci, B.A.

INVESTIGATORS:
Richard Wise, M.D., MRC. Path., Dudley Road Hospital, Dudley Road, Birmingham B18 7QH, England

Ages 24 to 40 years

ABSTRACT:
Aztreonam was administered as a single 2-min intravenous infusion of 1000 mg to 6 healthy male volunteers to investigate penetration of the monobactam into blister fluid. To assess the safety of aztreonam, clinical laboratory tests were conducted before and after drug administration.

Aztreonam was well-tolerated by all subjects, and no adverse reactions were apparent.

The pharmacokinetic profile of aztreonam was assessed by measuring aztreonam concentrations in multiple serum and urine samples from each subject after drug administration. In addition, multiple samples of blister fluid were collected from blisters formed by application of nine 0.2% cantharides plasters, 1x1 cm, to the anterior forearm of each subject approximately 12 hr prior to drug administration. All samples were assayed by the clinical investigator using a microbiological assay, and selected samples were subjected to high-pressure liquid chromatography assay. Both methods gave consistent results, in the judgment of the clinical investigator, and the bioassay data were chosen for detailed pharmacokinetic analysis.

The mean serum, blister fluid, and urinary concentrations of aztreonam obtained by bioassay are shown in Table I (following page). Aztreonam present at these concentrations would be expected to inhibit the majority of Enterobacteriaceae for approximately 8 hr in serum and blister fluid and 24 hr in urine, while Pseudomonas aeruginosa would be inhibited for approximately 4 to 6 hr in serum and blister fluid, and 24 hr in urine.

Based on the ratio, blister fluid AUC/serum AUC, aztreonam penetration into blister fluid averaged 70%.

The serum pharmacokinetics of aztreonam for individual subjects could be described by an open, linear, two-compartment kinetic model, which provided the mean pharmacokinetic parameters shown in Table III.

TABLE III[a]

Parameter	Mean ± S.E.M.
Distribution	
-Intercept	
V_1, liters/kg	0.13 ± 0.02
V_1^{ss}, liters/kg	0.20 ± 0.02
V_{area}^{ss}, liters/kg	0.23 ± 0.02
-Rate	
$t_{1/2\alpha}$, hr	0.38 ± 0.10
k_{12}, hr^{-1}	1.68 ± 1.18
k_{11}, hr^{-1}	1.70 ± 0.22
Elimination	
-Extent	
24-hr urinary recv., % of dose	74.6 ± 1.4
serum clear., ml/(min kg)	1.57 ± 0.08
renal clear., ml/(min kg)	1.04 ± 0.09
-Rate	
$t_{1/2\beta}$, hr	1.95 ± 0.11
k_{10}, hr^{-1}	1.00 ± 0.62

[a]Values are for 6 subjects.

The pharmacokinetic results shown in Table III for 2-min infusions of aztreonam agreed well with the results of pharmacokinetic studies of 2- to 30-min infusions of aztreonam (Protocols 18554-1, -2, -4, -8, and -18). The consistency between bioassay and HPLC assay results for aztreonam in the present study confirms similar findings reported previously and the conclusion that there were no detectable microbiologically active metabolites of aztreonam (Protocols 18554-2, -5, and -8).

TABLE I[a]

Time, hr	Concentration (µg/ml) Serum	Concentration (µg/ml) Blister Fluid
0.25	72 ± 5	—
0.5	54 ± 2	14 ± 4
1.0	42 ± 2	26 ± 4
4.0	11 ± 1	15 ± 2
6.0	5.3 ± 0.3	10 ± 1
8.0	3.6 ± 0.3	6.8 ± 1.2

Time, hr	Concentration (µg/ml) in Urine
0-2	1078 ± 445
2-4	333 ± 83
4-8	313 ± 123
8-12	112 ± 44
12-24	18 ± 10

Serum concentrations at 0.25, 1.5, 2, 3, 5 and 7 hours were recorded. Blister concentrations were recorded at 2, 3, 5 and 7 hours.

[a]All values are mean ± S.E.M. for 6 subjects, as determined by microbiological assay.

The maximum concentration (C_{max}), time to maximum concentration (T_{max}), and area under the concentration-time curve (AUC) for aztreonam in serum and blister fluid are shown in Table II.

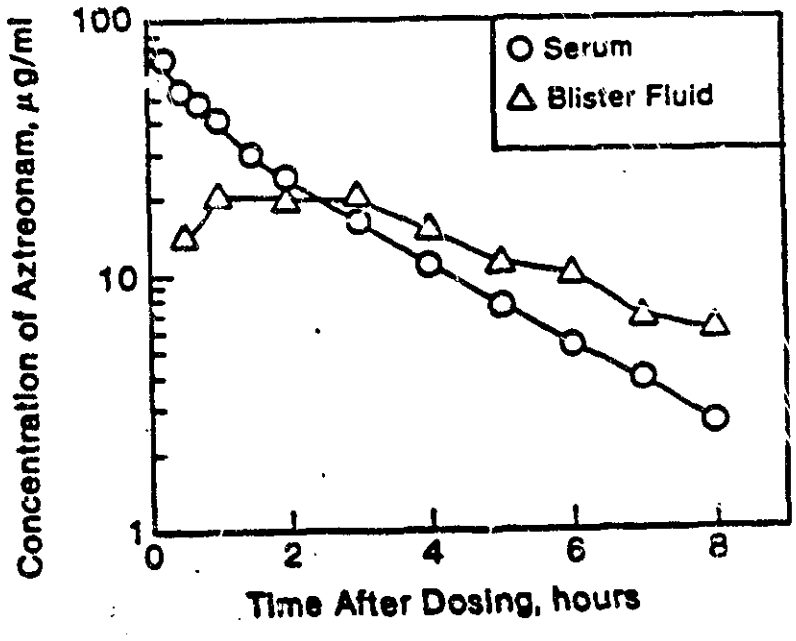
TABLE II[a]

Parameter	Serum	Blister Fluid	Blister Fluid/ Serum
C_{max}, µg/ml	73.5 ± 5.1	22.2 ± 3.1	—
T_{max}, hr	0.25 ± 0.00[b]	2.5 ± 0.8	—
AUC, µg × hr/ml	158.1 ± 15.8	107.4 ± 10.1	0.70 ± 0.05
$t_{1/2\beta}$, hr	1.95 ± 0.11	3.21 ± 0.15[c]	—

[a]Values are mean ± S.E.M. for 6 subjects.
[b]Time of first post-dose serum sample.
[c]Calculated by excluding Subject 4's $t_{1/2\beta}$ value of 46.2 hr, which was possibly influenced by poor blister formation.

FIGURE 1

PHARMACOKINETICS OF AZTREONAM IN SERUM AND BLISTER FLUID AFTER A 1000-MG 2-MIN INTRAVENOUS INFUSION IN SIX HEALTHY MALE SUBJECTS



Division of Medical Affairs

MNB-860

PRODUCT, ID NO. OR PROJECT NAME
SQ 26,776 (Aztreonam)

TITLE

Report on Addendum A to Protocol 18554-26 for Dr. P. O Madsen - A stud of Aztreonam Concentration in Human Prostate After A Single Intramuscular Dose

Study Protocol # 18,554-26

AUTHOR(S)

Lawrence T. Friedhoff, M.D., Ph.D.

INVESTIGATORS

Paul O. Madsen, M.D., Dept. of Urology, Univ. of Wisconsin School of Medicine
T.B. Platt, Ph.D. and R. Dhruv, Ph.D., Squibb Institute for Medical Research.

ABSTRACT

The purpose of this study was to determine serum, prostate and urinary bladder tissue aztreonam concentrations in patients who were given a single 1-gram intramuscular dose. Patients enrolled in protocol 18,554-26, "Comparison of Aztreonam with Placebo in Preventing Infection Following Transurethral Surgery", and randomized to receive aztreonam were selected for participation in this addendum study. Because of difficulty in obtaining tissue for assay, the stucy of bladder tissue was terminated after 3 specimens were obtained.

Patients scheduled for elective transurethral surgery were given a single intramuscular dose of aztreonam prior to the procedure. Serum, prostate and urinary bladder samples were obtained after dosing and assayed for aztreonam content by a microbiological binassay method. Samples of whole blood were also obtained and whole blood, prostate and bladder tissue were assayed for hemoglobin content. The results of the hemoglobin assays were used to correct total prostate and bladder aztreonam concentration for aztreonam present in tissue blood.

Eleven patients were enrolled in the addendum study. Patients ranged in age from 44 to 87 years (mean age 65.6 years), in height from 167.6 to 193.0 cm (mean 176.5 cm) and in weight from 61.4 to 101.2 kg (mean weight 81.4 kg). Eight patients had benign prostatic hypertrophy, one had prostate cancer and two had bladder cancer.

The results of the assays are shown in Table 1. The mean correction of prostate concentration for aztreonam in tissue blood was 6.9 percent (range 0 to 46 percent) of total tissue aztreonam. The mean time between dosing and serum sampling was 101 minutes (range 50-180 minutes). For serum samples taken simultaneously with prostate the mean concentration was 31.4 mcg/ml (range 18-46 mcg/ml).

The mean time between dosing and prostate sampling was also 101 minutes (range 50-180 minutes). The mean, uncorrected, prostate concentration was 7.78 mcg/gm (range 3.18 - 12.1 mcg/gm). The mean of the ratios of the uncorrected prostate to serum aztreonam concentrations was 0.25 (range 0.15-0.41)

Bladder specimens for assay of aztreonam content were obtained from three patients (see Table 1). The mean correction for aztreonam in tissue blood was less than 1 percent of total tissue aztreonam. Bladder tissue aztreonam concentration (uncorrected) was 10.6, 12.7 and 6.7 mcg/gm at 70, 77 and 125 minutes after dosing in these three patients.

A single 1-gram, intramuscular dose of aztreonam leads to prostate concentrations of between 3 and 12 mcg/gm. Although effectiveness cannot be demonstrated in the absence of a clinical trial, the concentrations of aztreonam attainable in prostate tissue are sufficiently high to suggest that aztreonam might be effective in the treatment of chronic prostatitis due to Enterobacteriaceae.

TABLE 1
Digester Results

Patient #	Preding Serum	Time of Sampling (minutes)	Prostate (mcg/gm) Corrected[a]	Prostate (mcg/gm) Uncorrected[a]	Serum (mcg/ml)	Prostate[b] Serum	Bladder Corrected[c]	Bladder Uncorrected[c]
001						-		-
002						0.22	-	-
003						0.41	-	-
004						0.24	-	-
005						0.21	-	-
006						0.15	-	-
007						0.27	10.6	10.6
008						-	12.7	12.7
009						0.22	-	-
010						0.19	-	-
011						0.30	-	-
MEAN	0	0	7.78	7.78	0	0.25	11.7	10.6

a - Not corrected for aztreonam in tissue blood

b - Correction for tissue blood was not possible because of small sample size.

c - Corrected for aztreonam in tissue blood.

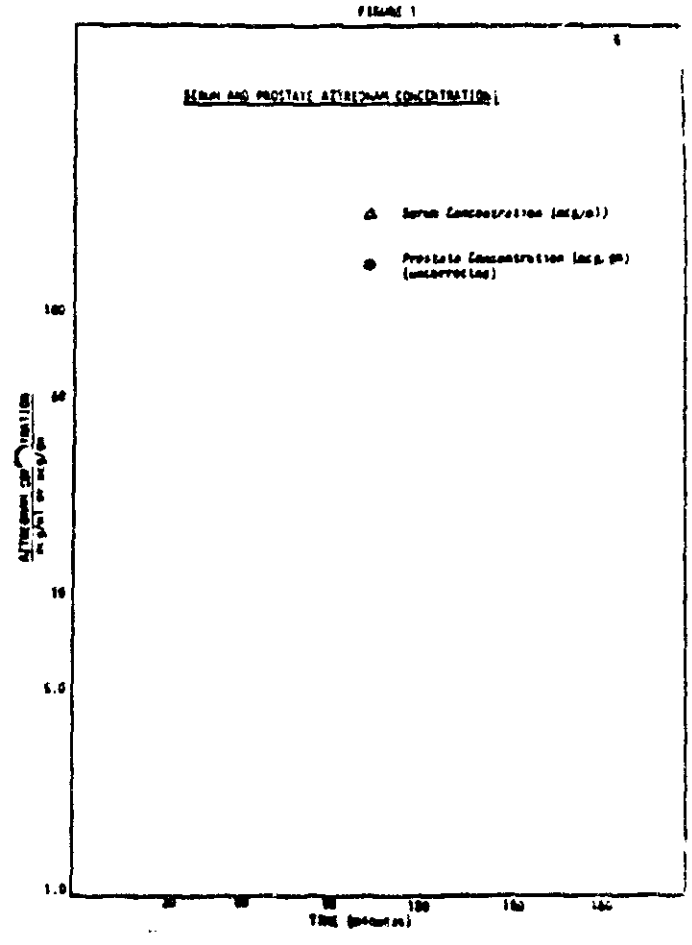
d - Time of serum.

e - Time of prostate.

f - Time of bladder.

g - For mean sampling times and serum concentrations see text.

FIGURE 1



Department of Clinical Pharmacology

SECTION
Division of Medical Affairs

PROJECT CODE
MNB-860

PRODUCT, SQ NO., OR PROJECT CODE
SQ 26,776 (Aztreonam)

TITLE
Report on Multiple Intravenous Dose Safety, Pharmacokinetic, and Bowel Flora Study of Aztreonam (SQ 26,776) in Patients with Cancer (30-Minute Infusions); Part A.

Study Protocol # 18,554-20, Part A

AUTHORS
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INVESTIGATORS
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T.B. Platt, Ph.D., The Squibb Institute.

ABSTRACT

Aztreonam was administered as 30-minute intravenous infusions of 1000 mg q8h for 2 to 15 days to 17 patients (9 evaluable patients were dosed for 6 to 9 days) undergoing cancer chemotherapy in laminar air flow rooms. To assess the effects of aztreonam on oral-pharyngeal and fecal flora, throat washings and fecal specimens were cultured on 2 separate days before and 2 days during aztreonam treatment. The pharmacokinetics of aztreonam and the metabolite SQ 26,992 were assessed by assaying multiple serum and urine specimens on 3 treatment days using microbiological and high-pressure liquid chromatography (HPLC) assays. To assess the safety of aztreonam, clinical laboratory tests were conducted before, during and after drug administration.

Patient 4 developed a maculopapular, pruritic rash over the trunk on Day 5. Aztreonam was discontinued, leading to resolution of the rash within 24 hours. No other possible adverse reactions to aztreonam were noted.

Patients 2, 3, 5 and 11 also left the study early after developing fevers due to suspected infections requiring antibiotic therapy. Patients 14, 15 and 17 provided specimens for kinetic studies on only 2 of the 3 prescribed treatment days. Endogenous flora and aztreonam pharmacokinetics were analyzed in the remaining 9 patients.

Although aztreonam had no significant effect on the numbers of oral aerobic bacteria and fungi, the monobactam produced a dramatic decline in fecal counts of aerobic gram-negative rods, without notable

Page 16 to bottom

Patients were not to have abnormal hepatic or renal function.

Note: mean steady-state AUC_{0-8} for Day 8 of study #18,554-4 in normal volunteers receiving aztreonam 1000 mg t.i.d. was 150.2 mcg hr/ml.

lterations in counts of other aerobic or anaerobic bacteria or fungi Table I). Aerobic gram-negative rods were initially present in the eces of 8 of the 9 evaluable patients, and were completely irradicated t the end of therapy in 5 of 8 (62.5%) patients.

TABLE I[a]

Source	Category of Microorganism	Pre	Pre	Day 3/5	Day 7/9
eces	Aerobic				
	Gram-Pos. Cocci	4.05×10^4	3.14×10^5	6.92×10^4	1.04×10^6
	Gram-Pos. Rods	8.36×10^3	2.78×10^3	1.67×10^3	1.29×10^2
	Gram-Neg. Cocci	0	0	0	0
	Gram-Neg. Rods	6.45×10^5	7.51×10^5	1.49×10^2	1.00×10^1
	Anaerobic				
	Gram-Pos. Cocci	4.64×10^0	0	0	7.82×10^0
	Gram-Pos. Rods	$3.8[illegible] \times 10^6$	6.74×10^6	8.00×10^7	6.07×10^0
	Gram-Neg. Cocci	0	2.76×10^1	0	7.74×10^0
	Gram-Neg. Rods	7.63×10^6	4.07×10^7	2.39×10^5	2.05×10^6
	Fungi	1.80×10^2	1.29×10^1	2.78×10^2	4.64×10^2
roat shings	Aerobic				
	Gram-Pos. Cocci	1.04×10^6	1.95×10^6	3.33×10^6	4.18×10^6
	Gram-Pos. Rods	8.37×10^4	3.10×10^4	2.99×10^3	3.23×10^3
	Gram-Neg. Cocci	3.93×10^1	4.27×10^1	6.23×10^1	1.41×10^2
	Gram-Neg. Rods	8.79×10^1	3.78×10^1	2.93×10^1	1.46×10^2
	Fungi	7.25×10^0	5.13×10^0	1.20×10^0	3.25×10^1

alues for feces are geometric mean colony counts per gram of feces, and for throat washings are geometric mean colony counts in the entire 20-ml throat ashing specimen; data from 9 patients.

here was close agreement between bioassay and HPLC assay results for ztreonam concentrations in serum and urine, indicating the lack of ioactive metabolites. For aztreonam, mean values for area under the erum concentration-time curve (AUC), maximum serum concentrations C_{max}), time to maximum serum concentration (T_{max}), elimination half-

ife ($t_{\frac{1}{2}}$), and 0-8 hr urinary excretion were not significantly different

n Days 1, 3 to 5, and 6 to 9 (Table II). In contrast, mean serum evels of SQ 26,992 rose from a mean value of 0.02 ug/ml at the onclusion of the first infusion of aztreonam on Day 1 to a steady-state evel of approximately 1 ug/ml on Days 3/5 and 6/9. Urinary excretion f SQ 26,992 was significantly lower on Day 1 compared to the later 2 ays when kinetic studies were done (Table II).

TABLE II[a]

Assay	Parameter	Day 1	Day 3/5	Day 6/9
Bioassay for Aztreonam	AUC_{0-8} (ugxhr/ml)	150.4 ±13.5	157.8 ±12.1	151.5 ±11.0
	C_{max} (ug/ml)	79.1 ±6.2	72.6 ±6.4	76.3 ±7.2
	T_{max} (hr)[b]	0.47 ±0.03	0.67 ±0.17	0.50 ±0.00
	$t_{\frac{1}{2}}$ (hr)	1.68 ±0.09	1.65 ±0.10	1.52 ±0.07
	Urin. Excr.[c] 0-8 hr (mg)	530 ±43	635 ±104	602 ±69
HPLC	Aztreonam Urin. Excr. 0-8 hr (mg)	414 ±54	596 ±113	549 ±69
	SQ 26,992 Urin. Excr. 0-8 hr (mg)	34.4[d] ±14.1	54.8 ±7.1	75.9 ±13.3

[a] All values are arithmetic mean ± SEM.

[b] Blood samples were drawn at 0.25, 0.5, 1, 2, and 4 hr after the start of a 30-min intravenous infusion; however, T_{max} was 0.25 hr for 1 patient on Day 1 and was 2.0 hr for another patient on Day 3/5, probably due to irregularities in the infusions.

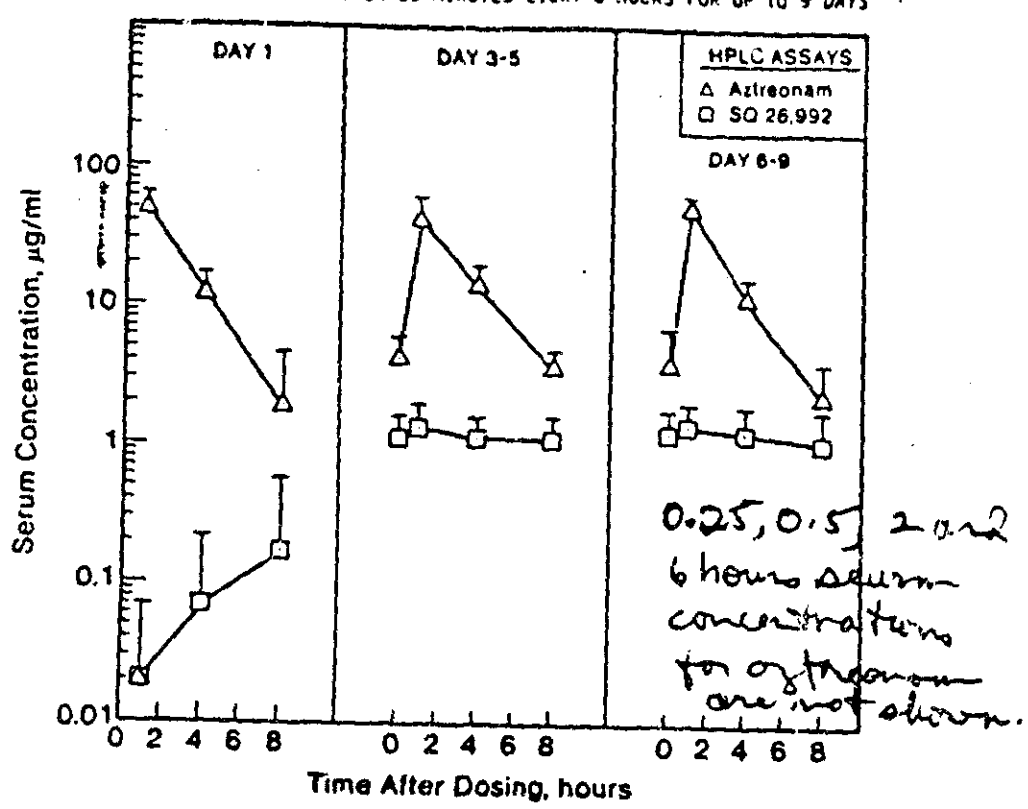
[c] The relatively low mean urinary recovery on Day 1 was probably due to incomplete 8-hr cumulative urine collections in several patients.

[d] Statistically significantly different from corresponding mean for Day 6 $P<0.05$.

Patient 5 died 10 days after discontinuation of aztreonam treatment due to an intracerebral hemorrhage and cardiac arrest. The clinical investigator judged that the cause of death was unrelated to the administration of aztreonam.

FIGURE 3

MEAN SERUM LEVELS OF AZTREONAM AND SQ 26,992 (HPLC ASSAY) IN CANCER PATIENTS RECEIVING 1 GRAM INTRAVENOUSLY OVER 30 MINUTES EVERY 8 HOURS FOR UP TO 9 DAYS



0.25, 0.5, 2 and 6 hours serum concentrations for aztreonam are not shown.

metabolite

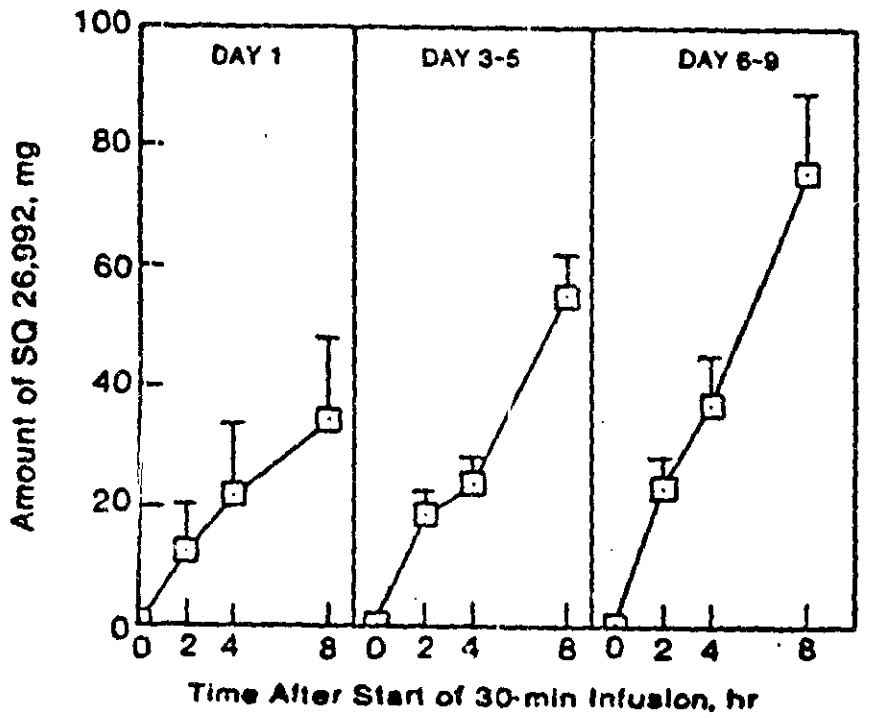
FIGURE 6

TABLE 3

CONCOMITANT MEDICATIONS

PATIENT NUMBER	CONCOMITANT MEDICATION	CONCOMITANT MEDICATION DOSE	CONCOMITANT MEDICATION UNITS
01	Prednisone	100	
	Amsacrine	77	
	Cytarabine	395	
02	Prednisone	600	
	Cytarabine	150	
03	Prednisone	100	
	Amsacrine	60	
	Cytarabine	140	
04	Prednisone	100	
	Amsacrine	50	
	Cytarabine	110	
05	Prednisone	100	
	Allopurinol	300	
	Amsacrine	67	
	Cytarabine	110	
06	Prednisone	60	
	Vincristine Sulfate	2	
07	Prednisone	100	
	Amsacrine	100	
	Cytarabine	120	
08	Prednisone	100	
	Allopurinol	300	
	Amsacrine	72	
	Cytarabine	210	
09	Cyclophosphamide	120	
	Etoposide	142	
	Daunorubicin	78	
	Theophylline	600	
	Cotrimoxazole Rx	100	
10	Amsacrine	67	
	Cytarabine	135	
11	Prednisone	100	
	Vitamins-combination	1	
	Cytarabine	100	
	Amsacrine Rx	9000	
12	Amsacrine	50	
	Cytarabine	130	
	Prednisone	100	
13	Asparaginase	35,000	
14	Cyclophosphamide	2700	
15	Prednisone	200	
	Amsacrine	52	
	Cytarabine	321	
16	Amsacrine	50	
	Cytarabine	125	

MEAN CUMULATIVE URINARY RECOVERY OF SQ 26,992 (HPLC ASSAY) IN CANCER PATIENTS RECEIVING 1 GRAM INTRAVENOUSLY OVER 30 MINUTES EVERY 8 HOURS FOR UP TO 9 DAYS



50-570 Vol. 8.1 Appndx III 11/12/85

DEPARTMENT:
Department of Clinical Pharmacology

SECTION:
Division of Medical Affairs

DATE OR PERIOD COVERED
September 30, 1984

PROJECT CODE
MNB-860

PRODUCT, EQ NO., OR PROJECT NAME
Aztreonam (SQ 26,776)

TITLE:
Report on Aztreonam (SQ 26,776) Penetration into Human Bone and Synovial Fluid after a Single Intravenous Dose.

AUTHORS:
Lawrence T. Friedhoff, M.D., Ph.D. and May Frantz, Ph.D.

INVESTIGATORS:
Catherine M. MacLeod, M.D. Rush-Presbyterian-St. Lukes Medical Center, Chicago, Illinois and Thomas B. Platt, Ph.D., The Squibb Institute for Medical Research, New Brunswick, NJ

ABSTRACT:

A single 2-gram dose of aztreonam (5-minute infusion) was administered preoperatively to 18 patients who underwent elective orthopedic (hip or knee) surgery. During the surgical procedure, specimens of cancellous bone and/or synovial fluid (and simultaneous serum) were obtained and assayed for aztreonam content. This use of aztreonam was safe and no definite adverse reactions were noted. Two patients had post-dose elevations of SGPT to 2.4 and 6.4 times normal. One of these patients also had an elevated post-dose SGOT (2.6 times normal). Two other patients had minor (less than twice normal) post-dose elevations of SGOT and SGPT and one of these also had a minor increase in LDH (1.2 times normal). These laboratory abnormalities were not associated with symptoms and required no treatment. The clinical investigator considered them possibly related to aztreonam.

Bone samples for assay of aztreonam were obtained at times from 0.98 to 2.09 hours after dosing (mean time 1.48 hours). Synovial fluid samples were obtained at times from 0.80 to 1.91 hours post-dose (mean time 1.24 hours). The results of the aztreonam assays are summarized in the following table. The mean bone and synovial fluid concentrations were 16.0 ug/gm and 83.0 ug/ml, respectively. The ratio of each individual bone or synovial fluid concentration to the simultaneously obtained serum concentration was also calculated. The mean ratios were 0.20 and 0.99 for bone and synovial fluid, respectively. Two patients had corrected bone concentrations of 0 ug/ml. These low values were probably due to over correction for aztreonam present in blood contained in these samples. The uncorrected bone aztreonam concentrations for these two patients were 6.8 and 5.2 ug/gm. The concentrations of aztreonam usually found in bone significantly exceeded the MIC_{90} for most *Enterobacteriaceae*, and the concentrations observed in synovial fluid significantly exceeded the MIC_{90} for most commonly encountered gram-negative organisms.

The high concentrations of aztreonam found in most bone and synovial fluid samples are consistent with the reported excellent therapeutic efficacy of aztreonam in patients with osteomyelitis and infectious arthritis caused by gram-negative organisms. These concentrations also suggest that aztreonam might be useful as a prophylactic agent for patients who have a high risk of gram-negative contamination of bone or synovial fluid.

AZTREONAM CONCENTRATIONS CORRECTED FOR AZTREONAM IN BLOOD

(ug/gm or ug/ml)

Pat. No.	Pre-Dose Serum	0.5-hour Serum	Serum Simult.w/Bone or Synovial Fluid	Bone[a]	Synovial[b] Fluid
1					
2					
3					
4					
5					
6[d]					
7					
8					
9					
10					
11					
12					
13					
14					
15					
16					
17					
18					
Mean	0	124.0	77.8	16.0	83.0
SEM		8.9	5.1	4.3	9.2
(n)		(16)	(17)	(15)	(11)

[a] Femoral head.
[b] Sample of patient 7 was from the knee joint, all other samples were from the hip joint.
[c] Sample potency was greater than 88.8 ug/ml; however, because of the small volume, the exact potency could not be determined.
[d] Adequate samples could not be obtained from patient 6.
[e] The pre-dose sample was accidentally taken after the injection of aztreonam was begun. The assayed potency was 33.9 ug/ml.
[f] No sample could be obtained at 0.5 hours. The 1.91-hour serum concen-

TISSUE STABILITY AND INTRARENAL DISTRIBUTION OF AZTREONAM IN HEALTH AND DISEASE

A. J. WATSON, R. L. STOUT, D. K. SOOD, A. MILTON

The new monocyclic beta-lactam aztreonam is emerging as a clinically important and potent beta-lactamase resistant antibiotic for the management of all aerobic gram-negative infections. Our investigations were designed to: 1) Quantify the long-term stability of the drug in renal tissues; 2) Characterize the intrarenal distribution of the drug in a healthy canine experimental animal model subjected to the physiologic variables of dilute urine production (hydrated state n=4), concentrated urine production (hydropenic state n=5), acid urine production (pretreatment with ammonium chloride n=3), and alkaline urine production (sodium bicarbonate administration n=4); and 3) Measure tissue concentrations achievable with aztreonam in severely diseased human renal tissues. All plasma, urine and tissue assays for aztreonam content were performed utilizing an agar well diffusion microbiologic assay system.

Renal cortical, medullary and papillary tissues from healthy non-drug treated dogs were homogenized in pH 6.0 phosphate buffer and known amounts of aztreonam, spanning the anticipated tissue drug concentration range, were added to the homogenate. Multiple tissue aliquots were stored at -20°C and -70°C. The -70°C tissues were then assayed for residual drug activity at 2-4 weeks for a period of 12 weeks. Full recovery of drug activity was noted in all these tissue samples. Tissues stored at -20°C and assayed 12 weeks later showed significant reduction in renal cortical activity with retention of 50% to 75% of drug activity in medullary and papillary tissues respectively. Fresh renal tissues were harvested from aztreonam treated dogs and stored at -70°C for twelve weeks or more prior to homogenization in preparation for a microbiologic assay there was a near total loss of drug activity in renal cortical tissues.

Sixteen dogs were incorporated in our studies defining the influence of renal physiologic parameters upon intrarenal aztreonam distribution. The following summary table presents mean (±S.E.) plasma, tissue, and urine concentration data together with renal clearance values, urine flow rate and urine pH result for the drug.

PHYSIOLOGIC STATE	PLASMA µg/ml	CORTEX µg/gm	MEDULLA µg/gm	PAPILLA µg/gm	URINE µg/ml	pH	F.O.A ml/min
Hydrated	22±1.3	5.5±0.7	34±3	50±7	383±40	6.9	.65±.1
Hydropenia	21±2.1	5.4±1.7	27±3.5	52±5	1279±199	7.4	.29±.0
Acid Urine	28±4	7.5±1.7	38±5	76±5	4646±651	6.1	.18±.0
Alkaline Urine	26±2	7.2±2.9	30±5	58±7	1020±109	8.1	.45±.2

The results indicate that during all prevailing renal physiologic circumstances produced in these studies aztreonam manifests a significant 8 to 10-fold increase in drug concentration from the renal cortex to the papilla. Tissue levels were not markedly influenced by the state of hydration or urine ph. However, urine concentrations of the drug were significantly increased during the production of concentrated urine. The presence of acid urine production further increased urine drug concentration and clearance rates.

The concentration of aztreonam achievable in severely diseased human renal tissues were determined in two patients undergoing nephrectomy in preparation for renal transplantation. In these diseased tissues aztreonam levels were 5-6µg/gm tissue and 30-62µg/gm tissue, values that were similar to or less than the concomitant plasma concentrations but nonetheless substantially greater than the MIC values of the typical gram-negative pathogen for which this drug may be used in complicated upper and lower urinary tract infections.

In bacterial pyelonephritis it is within the medullary and papillary zones of the kidney that acute seeding and chronic replication of bacterial activity takes place. In view of the high concentrations of aztreonam noted in the latter zones of the kidney it will be of therapeutic importance to undertake further studies designed to solidly identify a potential clinical therapeutic correlation between the high aztreonam levels achievable in the inner zones of the kidney and eradication of bacterial pyelonephritis.

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IV.

M E M O R A N D U M

DEPARTMENT OF HEALTH & HUMAN SERVICES
Public Health Service
Food and Drug Administration
Center for Drugs and Biologics
Office of Drug Standards

DATE : April 30, 1986

TO : Dr. Francis Min
Division of Anti-Infective Drug Products (HFN-815)

THROUGH: Acting Chief, Pharmacokinetics Evaluation Branch (HFN-226) CTV 5/9/86

FROM : Acting Section Head
Pharmacokinetics Evaluation Branch (HFN-226)

SUBJECT: NDA 50-580 - Aztreonam Injection (E. R. Squibb), Preliminary NDA Review Addressing Package Insert Dosing Recommendations for Renal Impaired Patients

Forwarded for your evaluation is a portion of the Division of Biopharmaceutics (DB) NDA review for aztreonam that will be coming to completion in the near future.

Knowing that your Division is also near completion in its review of this application, we are forwarding this portion of the DB review at this time so that the items and issues that are raised can be further addressed, if necessary, before our review is formally completed.

John P. Hunt
Division of Biopharmaceutics

Attachment

cc: HFN-815(Tabor), HFN-225(Hunt, Viswanathan), Chron, Drug

IV.

COMMENTS ADDRESSING AZTREONAM DOSE ADJUSTMENTS IN RENAL IMPAIRED PATIENTS

The following comments address within and between study comparisons (Study Nos. 18,554-8 and 24; Addenda to Study Nos. 18,554-27 and 31) as related to dose adjustments for aztreonam that are proposed in the package insert for patients with decreased renal function.

1. The proposed package insert indicates the following dose adjustments for renal dysfunction for aztreonam:

Renal Impairment

Prolonged serum levels of aztreonam may occur in patients with transient or persistent renal insufficiency. Therefore, the dosage of AZACTAM should be halved in patients with estimated creatinine clearances between 10 and 30 mL/min/1.73 m^2 after an initial loading dose of 1 g or 2 g.

When only the serum creatinine concentration is available, the following formula (based on sex, weight, and age of the patient) may be used to approximate the creatinine clearance (Clcr). The serum creatinine should represent a steady state of renal function.

$$\text{Males: } Clcr = \frac{\text{Weight (kg)} \times (140 - \text{age})}{72 \times \text{serum creatinine (mg/dL)}}$$

Females: 0.85 x above value

In patients with severe renal failure (creatinine clearance less than 10 mL/min/1.73 m^2), such as those supported by hemodialysis, the usual dose of 500 mg, 1 g or 2 g should be given initially. The maintenance dose should be one-fourth of the usual initial dose given at ~~fixed intervals of 6, 8 or 12~~ the usual fixed interval hours. For serious or life-threatening infections, in addition to the maintenance doses, one-eighth of the initial dose should be given after each hemodialysis session.

Dosage in The Elderly

Renal status is a major determinant of dosage in the elderly. These patients in particular may have diminished renal function; serum creatinine may not be an accurate determinant of renal status. Therefore, estimates of creatinine clearance should be obtained, and appropriate dosage modifications made if necessary.

NOTE: Appendix I gives the Dosage and Administration section of the package insert as filed on 2/12/86.

2. The above proposed aztreonam dose adjustments for renal insufficiency, as selected by Squibb for its package insert, appear to be based in part (i.e. for severe renal failure) upon the results of two studies (#18,554-8 and 18,554-24) where single 2 minute intravenous infusions of aztreonam were given to patients with varying degrees of renal dysfunction. Attachment I gives the health status for each patient enrolled in those studies.

Based upon the independent findings from the two separate single dose studies, the investigators of those studies proposed the same drug dose adjustments using a constant dosage interval approach as follows (i.e., using urinary creatinine clearance vs. drug serum clearance analyses; Figures 1 and 2).

		Creatinine Clearance (ml/min) Grouping				
Study No.	No. of Patients:	8	5	5	6	
18,554-8		83-164[1]	30-54	11-28	0-2 Not Req. HD.[2]	Req. H.D.
18,554-24	No. of Patients:	5	5	6	4	5
		91-137	35-61	13-24	4-9	anuric
Clcr[3] Ranges For Dose Adjustment Investigators' Recommended Dose Adjustments (Fraction of Normal Dose)		> 80 No Adjustment	30-80 1/2*	10-29 1/3*		< 10 1/4*

1 Actual range of determined creatinine clearance values.
2 H. D. = hemodialysis.
3 Cl_{cr} = creatinine clearance (ml/min).
*All patients with renal insufficiency are to receive a loading dose of the drug equal to the dose used in patients with normal renal function (>80ml/min).

3. In support of their proposed dose adjustments, the study investigators for Study No. 18,554-8 made predictions of what steady-state aztreonam serum levels would be achieved using their recommended dose adjustments for renal dysfunction (e.g., Figure 10). Example, for t.i.d. dosing the predicted trough serum levels ranged between 3 mcg/ml and 8.2 mcg/ml depending the Clcr group and dose administered.

Advantages and disadvantages given in the NDA regarding the proposed dose adjustments that were determined from Study 18,554-8, using the constant dosage interval approach, are as follows.

The biological half-life of aztreonam is about 1.7 hours when renal function is normal, and about 6 hours in severe renal insufficiency (Table 49). Therefore, a multiple-dose regimen will reach steady-state conditions in approximately 5 x 1.7 = 8.5 hours in patients with normal renal function, and in approximately 5 x 6 = 30 hours in patients with severe renal insufficiency. Patients with severe renal insufficiency can be maintained on less than the normal aztreonam dose (Table 57); however, a loading dose will be necessary to avoid a delay in time before the serum concentration reaches steady-state (Chennavasin and Brater, 1981). This is particularly important, because severe gram-negative infections can be rapidly fatal, unless appropriate antibiotics are administered promptly and in adequate doses. The recommended loading dose of aztreonam would be identical to the dose used in patients with normal renal function.

This dosing method has the following advantages: 1) the average serum concentration of aztreonam is the same for various degrees of renal insufficiency, and 2) the standard dosage interval is kept the same. Disadvantages include the following: 1) this dosing method allows peak serum levels to fall, possibly resulting in a shorter duration of bactericidal activity in patients with bacteria for which the minimum bactericidal concentrations for aztreonam are high (Dettli, 1977), 2) odd doses may predispose to medication errors, 3) increased minimum serum levels are thought to be a risk factor for drug toxicity (Chennavasin and Brater, 1981), and 4) a loading dose is necessary to reach steady-state conditions quickly.

4. If the predicted t.i.d. dosing steady-state drug serum levels from Study No. 18,554-8 are in fact the clinically desirable levels (Figure 10), then 1) there are discrepancies between some of the recommended aztreonam doses proposed by the investigators of Study Nos. 18,554-8 and 24 and the doses actually proposed in the package insert and 2) there are discrepancies between predicted observed drug levels that were obtained using the labeling's recommended dose adjustments (see Comment #5).

a. The investigators of Study Nos. 18,544-8 and 24 recommend that for patients with Clcr values between 10 to 30 ml/min/1.73m^2 only 1/3 the normal dose that would be given to patients with normal renal function should be administered. On the other hand, the package insert recommends that 1/2 of the normal dose (i.e., more drug) should be given.

b. Additionally, it should be noted that there are no dose adjustments recommended in the package insert for patients with Cl_{cr} between 30-80ml/min. The investigators of Study Nos. 18,544-8 and 24 recommend the dose should be reduced by 1/2 for patients in the Clcr range of 30-80 ml/min (Note: Actual observed Cl_{cr} values ranged between 30 and 61 ml/min). The importance of these points from a clinical perspective should be reviewed in conjunction with Comment #5 below.

5. In Addenda to Study Nos. 18,554-27 and 31, presented were aztreonam and SQ 26, 992 (major metabolite) serum trough levels for nine patients with different degrees of renal dysfunction (Figures 1A and 2A). Drug was intravenously administered to the patients under multiple dose conditions (t.i.d.) for 5 to 10 days (see Attachment II). According to the sponsor, Patients Nos. 7 and 9 died following termination of drug therapy (see Attachment III) but the deaths were judged to be unrelated to aztreonam administration.

The points addressed below (i.e., items a and b) address high trough drug levels as related to possible drug toxicity as might be as suggested in Comment No. 8 above.

a). In Study Nos. 18,554-27 and 31, patient Nos. 2 (70.5 kg), 5 (80.4 kg), 6 (63.4 kg), and 7 (66.5 kg) (Clcr = 13 - 28ml/min) received t.i.d. doses of aztreonam as might occur according to the dose adjustments as proposed in the package insert for patients with Cl_{cr} between 10-30 ml/min/1.73^2 (i.e., 1/2 the normal dose given to patients with normal renal function). For these patients, observed mean trough drug levels (Table 5, Attachment II) were 25.9, 42, 30 and 30.8 mcg/ml, respectively.

Comparison of these observed mean steady-state trough levels of aztreonam with those predicted using the analysis procedures of Study No. 18,554-8 indicate that the observed mean trough concentrations are "about" 2.3, 3.8, 2.7 and 1.4 times greater than would have been predicted for Patients Nos. 2, 5, 6 and 7, receiving t.i.d. doses of 0.5 or 1.0 doses respectively. Similarly, and maybe more importantly, are the findings for Patient No. 9 (Cl_{cr} = 5 ml/min). This patient (76.6 kg) received a dosing regimen that could be given as is proposed in the package insert (i.e., 500 mg t.i.d.). Observed mean (range) aztreonam trough levels were 58.9 (41.3 to 71.8) mcg/ml. The observed mean trough level was approximately 3.6 times greater than what would have been predicted using the data analysis approach of Study No. 18,554-8 for a patient with a Cl_{cr} = 5 ml/min.

Additionally, as a point of interest are the findings for Patient No. 1 (31.7 kg) who had a calculated Cl_{cr} of 66 ml/min and a mean (range) aztreonam trough value of 61.7 [redacted] mcg/ml following a 1 g t.i.d. dosing schedule. If, in fact, high drug trough levels are a concern as related to potential drug toxicity as might be suggested in Comment No. 3 above, then according to the current package insert, no dose adjustment would have been required for this patient based upon his creatinine clearance value. The investigators for Study Nos. 18,554-8 and 24 would have recommended a dose reduction by 1/2 for a patient with a Clcr in this range.

b) Similarly, as concerns might be raised over the levels of aztreonam achieved in renally impaired patients, concerns might also be raised over the levels of major metabolite (SQ 26, 992) achieved under the same conditions. Example, Patient 9 reached metabolite serum trough levels of [redacted] and [redacted] mcg/ml on treatment days 7 and 8 (Table 5, Attachment II; Figure 2A).

Raised can be questions about the potential toxicity of this major metabolite under multiple dose conditions, especially in renal impaired patients.

6. Summary Comments

Knowing that two patients (#7 and 9) who had renal impairment died following aztreonam treatment in Study Nos. 18,554-27 and 31, the results and data analyses of Study Nos. 18,554-8, 24, 27 and 31 were reviewed critically from a pharmacokinetic perspective. Taking into account the points discussed in Comments #1-5 above, the following should be brought to the attention of the reviewing Medical Officer for consideration as related to the proposed aztreonam dose adjustments for renal impairment in the package insert.

a) For aztreonam, if there are safety/toxicity concerns over having persistently high drug serum trough levels (e.g.. in some cases as high as 70 mcg/ml) as is alluted to in Comment #3, then the proposed dose adjustments as given in the package insert are likely to result in drug trough serum levels that will be higher than predicted using those dosing adjustments. The assumption here being that the predicted t.i.d. drug levels from Study No. 18,554-8 are the clinically desirable drug levels to be maintained. Based upon this, should more conservative dose adjustments be used than are currently recommended in the labeling for renal impaired patients?

b) If high aztreonam drug levels are of concern in renal impairment, should dose adjustments also be recommended for patients with Cl_{cr} greater than 30ml/min but less than 80 ml/min (or 60 ml/min). The investigators of Study Nos. 18,554-8 and 24 recommend dose adjustment in this Cl_{cr} range but the current package insert does not.

c) For the major metabolite of aztreonam are there concerns over its potential accumulation and its potential for toxic effects? From Study Nos. 18,554-27 and 37, increasing major metabolite trough serum levels were observed in treated patients. In normal volunteers major metabolite serum levels were only negligible.

d) Although the patients (#7 and 9) that died, as reported in Study Nos. 18,554-27 and 31, may have had cause of death unrelated with drug treatment it should be noted that mean (range) trough drug levels were 30.8 ([redacted] mcg/ml and 58.9 ([redacted] mcg/ml, respectively.

[redacted]

study # 18554-8

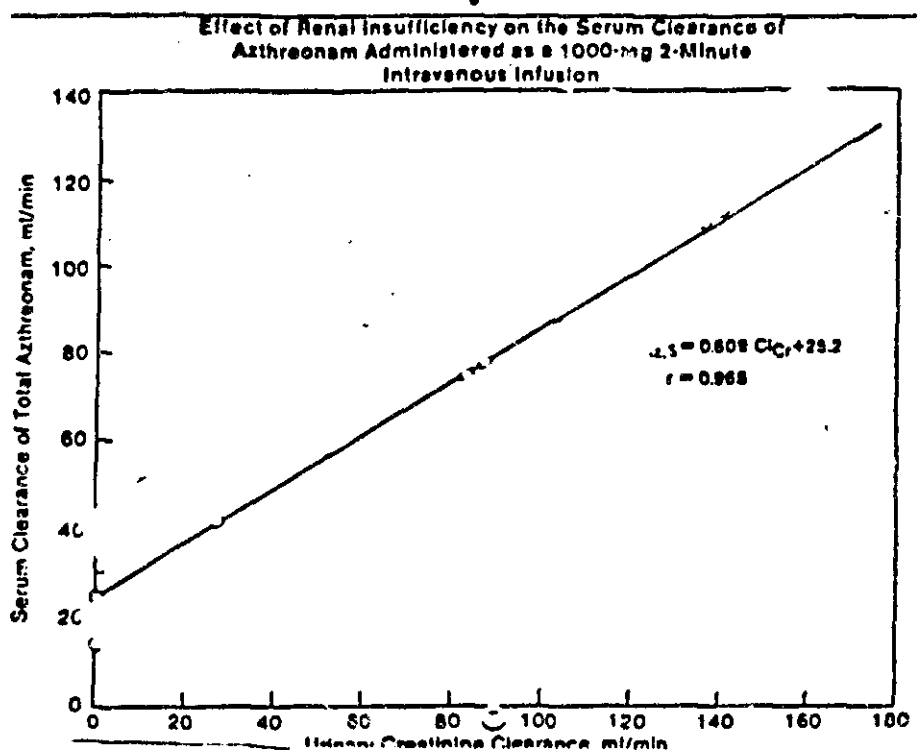


Figure 2
study # 18554-24

Effect of Renal Insufficiency on Serum Clearance (HPLC Assay) of Aztreonam Administered as a 1000-mg 2-Minute Intravenous Infusion

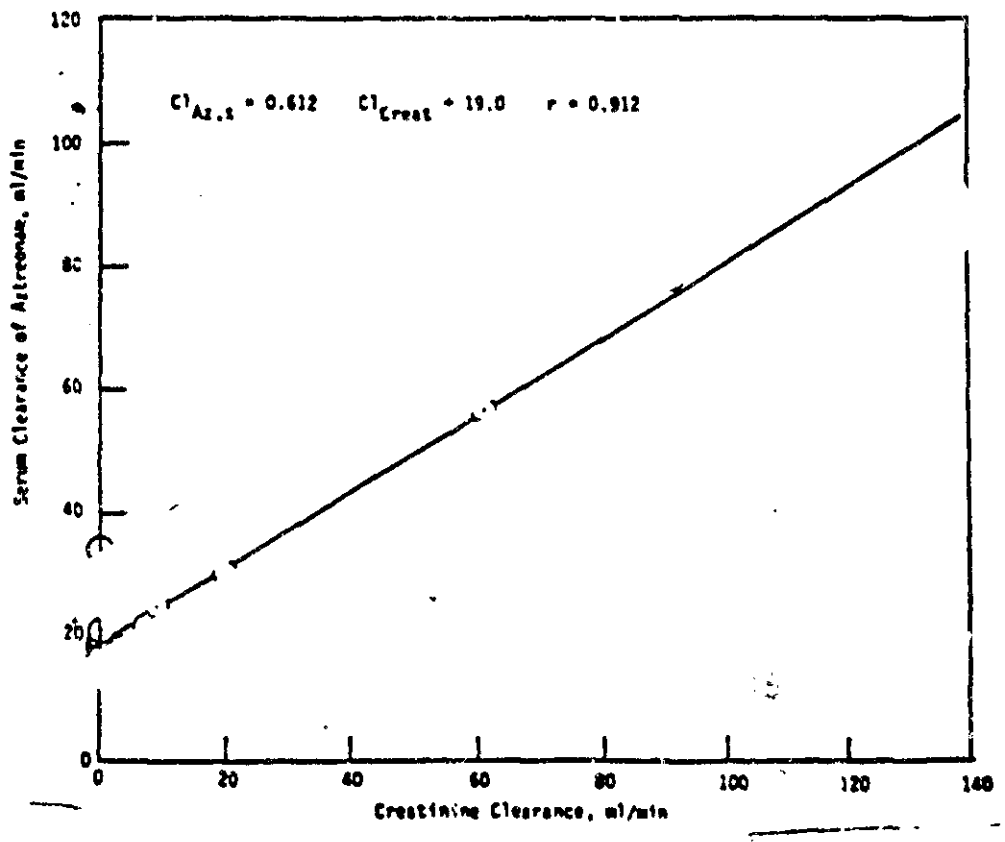
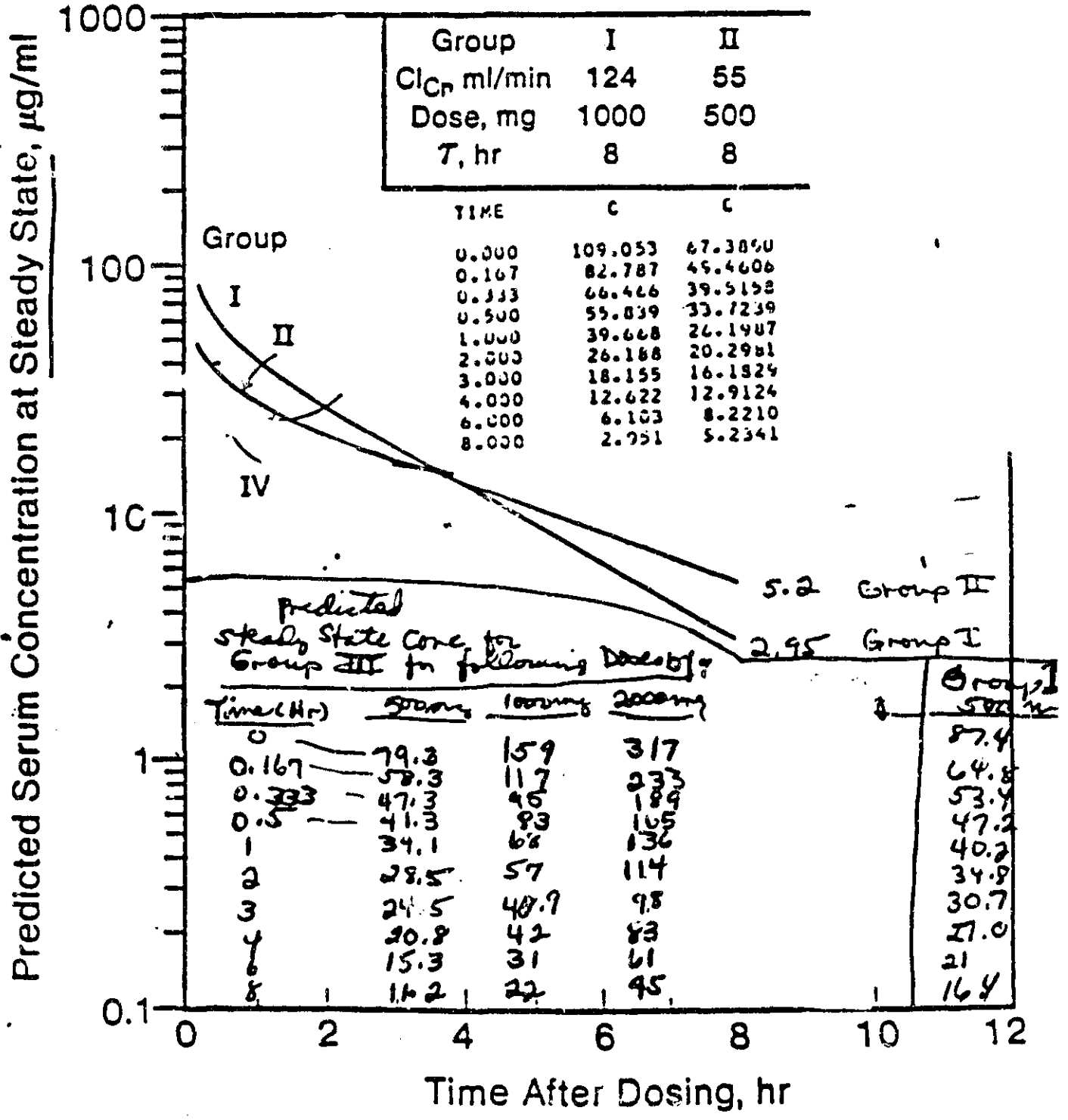


FIGURE 10

Predicted Serum Azthreonam Concentrations for a Dosage Regimen Modified for Renal Insufficiency: Constant Dosage Interval (8 Hours) and Variable Dose (Beginning at 1000 mg)



Group	I	II
Cl_{Cr} ml/min	124	55
Dose, mg	1000	500
T, hr	8	8

TIME	C	C
0.000	109.053	67.3850
0.167	82.787	45.4606
0.333	66.466	39.5158
0.500	55.839	33.7239
1.000	39.668	26.1907
2.000	26.188	20.2981
3.000	18.155	16.1824
4.000	12.622	12.9124
6.000	6.103	8.2210
8.000	2.951	5.2341

SQUIBB RESEARCH AND DEVELOPMENT
DIVISION OF MEDICAL AFFAIRS

Attachment I

Study #1855-4-8

TABLE 2

MEDICAL HISTORY AND PHYSICAL FINDINGS*

GROUP NUMBER	PATIENT NUMBER		PERTINENT MEDICAL HISTORY AND PHYSICAL FINDINGS	Kg
I	113	(122) 04	33 Aortic Incompetence Bicuspid Aortic Valve	74
	73	(87) 06	39 Hypertension Gout	83
	80	(83) 12	56 Hem. Glomerulonephritis Coronary Artery Bypass Graft	92.3
II	55	(54) 07	46 Hypertension Polycystic Kidneys	82.1
	29	(31) 09	52 Hypertension COPD Gout Glomerulonephritis	87.3
	43	(51) 11	35 Hypertension Glomerulonephritis	95
	26	(40) 13	34 Hypertension Membranous Glomerulopathy	96.4
	29	(48) 15	39 Hypertension Glomerulonephritis	63.3

*Healthy volunteers are not listed.

TABLE 2 (continued)

GROUP NUMBER	PATIENT NUMBER		PERTINENT MEDICAL HISTORY AND PHYSICAL FINDINGS	Kg
III	33	(29) 10	30 Hypertension Hypertensive nephrosclerosis	83.6
	24	(31) 14	41 Nephrosclerosis Mental Retardation Diabetes Mellitus	83.5
	23	(27) 16	64 Hypertension Steroid Arthropathy Hips Steroid Arthropathy Knee Mild Psoriasis Chronic Interstitial Nephritis	93.8
	20	(27) 17	50 Hypertension Peripheral Vascular Disease Diabetes Mellitus Hypertensive and Diabetic Nephropathy	96.8
	14	(17) 18	34 Hypertension Glomerulosclerosis	83.2
IV	0	0 19	35 Hypertension Renal Osteodystrophy Gouty Arthritis Hypertensive nephrosclerosis	78.8
	0	0 20	29 Gouty Hereditary Nephropathy	63.2
	0	0 21	28 Chronic Renal Failure ← Gouty Arthritis Hereditary Nephropathy	70.5
	0	0 22	45 Hypertension Renal Osteodystrophy Hypertensive nephrosclerosis	80
	0	2 23	50 Hypertension Nephrosclerosis	83.1
	3	1 24	46 Polycystic Kidneys End Stage Renal Disease	82.6

Protocol 1855-4-8

TABLE II

MEDICAL HISTORY AND PHYSICAL FINDINGS*

GROUP NUMBER	PATIENT NUMBER		PERTINENT MEDICAL HISTORY AND PHYSICAL FINDINGS	Kg
II	35	06 61	Renal Parenchymal Disease Chronic Pyelonephritis Arterial Hypertension Coronary Disease Cardiac Ischemia Tuberculosis Nephrectomy	69.6
	61	07 37	Chronic Glomerulonephritis Hematuria	65.1
	41	08 50	Chronic Pyelonephritis Hypertension	76.6
	39	09 38	Hypertensive Renal Disease Renal Parenchymal Disease Arterial Hypertension Polycystic Kidneys	55.6
	61	10 32	Chronic Pyelonephritis Arterial Hypertension	63
III	21	11 65	Chronic Pyelonephritis Arterial Hypertension Duodenal Ulcer	67.3
	23	12 31	Hypertensive Renal Disease Chronic Glomerulonephritis Arterial Hypertension Left Ventricular Hypertrophy	59.6
	20	13 56	Renal Parenchymal Disease Arterial Hypertension Polycystic Kidneys	77.3
	13	14 34	Chronic Glomerulonephritis Arterial Hypertension	72.3
	24	15 63	Chronic Glomerulonephritis Arterial Hypertension	54.0
	13	16 66	Chronic Pyelonephritis Arterial Hypertension Gout	74.4

*Healthy volunteers are not listed.

TABLE II (Continued)

MEDICAL HISTORY AND PHYSICAL FINDINGS

GROUP NUMBER	PATIENT NUMBER		PERTINENT MEDICAL HISTORY AND PHYSICAL FINDINGS	Kg
IV	9	16 65	Polycystic Disease of Kidneys Arterial Hypertension Anorexia, Nausea Large Kidneys	61.1
	4	17 62	Chronic Pyelonephritis Pneumonitis	41
	9	19 52	Polycystic Disease Arterial Hypertension Coronary Disease Chronic Respiratory Failure Polyneuritis	6.6
	8	20 65	Respiratory Insufficiency Chronic Pyelonephritis Prostate Adenoma	63.9
	Anuric	21 46	Hemodialysis Chronic Pyelonephritis Chronic Pericarditis Tuberculosis Ureterostomy	54
	Anuric	22 56	Hemodialysis Renal Parenchymal Disease	93.8
	Anuric	23 60	Hemodialysis Renal Parenchymal Disease Polycystic Disease Arthralgia	59
	Anuric	24 61	Hemodialysis Hypertensive Renal Disease Hypertension	51.4
	Anuric	25 25	Renal Failure Hemodialysis Chronic Glomerulonephritis	62.6

Investigators: F.R. Sattler, M.D., Hershey Medical Center, Penn State University, Hershey, PA.

Pharmacokinetics of aztreonam in patients with urinary tract infections and renal insufficiency (Addenda A to Protocols 18554-27 and -31)

Serum trough concentrations (levels in serum obtained just prior to a scheduled dose) of aztreonam and SQ 26,992 were measured daily in 9 patients (2 enrolled in Protocol 18554-27 and 7 enrolled in Protocol 18554-31) with renal insufficiency. These patients were receiving 500, 1000, or 2000 mg aztreonam intravenously q8h x 5 to 10 days for treatment of serious urinary tract infections. One patient received hemodialysis and provided additional blood specimens before and after hemodialysis. Serum concentrations of aztreonam and SQ 26,992 (the major metabolite of aztreonam resulting from hydrolytic opening of the beta-lactam ring) were measured in all patients by high-pressure liquid chromatography.

The 5 male and 4 female Caucasian patients enrolled in these addenda studies had ages ranging from 56 to 83 years (mean, 72 years) and body weights from 49.5 to 81.7 kg (mean 67.7 kg).

Aztreonam trough levels were generally stable in the 20 to 70 µg/ml range during treatment. Such levels would be potentially therapeutic for commonly encountered *Enterobacteriaceae* and *Pseudomonas aeruginosa*. SQ 26,992 concentrations tended to increase to steady-state levels during therapy in proportion to the degree of renal insufficiency. Initial serum levels of SQ 26,992 were in the 1.9 to 5.0 µg/ml range, and Day 5 or 6 levels were in the 3.0 to 29.3 µg/ml range. The ratio of Day 5 or 6 serum SQ 26,992 concentration to the initial serum level (before the third or fourth dose on Day 1 or 2) was near unity in patients with creatinine clearances in the 25 to 40 ml/min range, and was between 3.5 and 11.3 in patients with creatinine clearances below 25 ml/min. Aztreonam and SQ 26,992 underwent hemodialysis; nevertheless, the highest serum levels of SQ 26,992 were measured in a patient being supported on chronic hemodialysis.

Aztreonam was tolerated well by patients with serious urinary tract infections. A possible drug-related adverse effect in 1 of 9 patients consisted of mildly elevated serum transaminases. Thus, the accumulation of SQ 26,992 had no clinically important effects.

TABLE 1

DEMOGRAPHIC DATA AND DOSAGE REGIMENS

Addendum Patient No. (Initials)	Data Protocol No. 18554-	Pt. No.	Age	Sex	Height (cm)	Wt., (kg)	Aztreonam Dose (IV)
[redacted]	-31	5	66	M	[redacted]	81.7	1gm q8h x 5d
[redacted]	-31	7	75	F	162.0	70.5	0.5gm q8h x 6d
[redacted]	-27	31	75	F	160.5	60.4	2gm q8h x 10d
[redacted]	-27	32	83	F	152.8	49.5	2gm q8h x 5d
[redacted]	-31	9	66	F	171.8	80.6	1gm loading, 0.5gm q8h x 6d
[redacted]	-31	10	70	M	174.0	63.4	1gm loading, 0.5gm q8h x 7d
[redacted]	-31	12	81	M	[redacted]	66.5	2gm loading, 1gm q8h x 8d
[redacted]	-31	14	80	M	[redacted]	68.8	1gm q8h x 8d
[redacted]	-31	8	62	M	[redacted]	76.6	1gm loading, 0.5gm q8h x 6d
Mean (Range)			72 (56-83)			67.7 (49.5-81.7)	

* Calculated using Cockcroft-Gault et al, 1976.

TABLE 1 (cont.)

SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992

Add. Pt. No.	4		5		6	
Treatment	AZ	SQ 26,992	AZ	SQ 26,992	AZ	SQ 26,992
Day						
1	48.0*	5.56*	[redacted]	[redacted]	[redacted]	[redacted]
2	41.6	5.28	43.0*	5.04*	32.1*	2.31*
3	40.5	5.33	35.7	5.94	31.8	6.43
4	40.8	4.90	41.0	13.8	28.8	7.83
5	36.9*	4.63*	45.5	17.9	32.1	8.24
6	40.0	5.24	42.8*	20.8*	27.5*	8.21*
7	42.0	4.98	[redacted]	[redacted]	[redacted]	[redacted]
8	[redacted]	[redacted]				
9						
10						
11						
12						
13						
14						
Area Under Serum Conc. Day 5 or 6 to Day 1 or 2	41.4	5.02	42.0	10.0	30.0	6.47

TABLE 1

SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992

Add. Pt. No.	1		2		3	
Treatment	AZ	SQ 26,992	AZ	SQ 26,992	AZ	SQ 26,992
Day						
1	30.7*	1.93*	[redacted]	[redacted]	[redacted]	[redacted]
2	66.7	8.57	28.1*	2.32*	27.1*	3.03*
3	57.8	8.42	37.1	8.84	26.6	3.61
4	60.8	16.2	26.0	12.8	19.5	3.15
5	57.7*	16.9*	21.8	14.8	22.7*	2.99*
6	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
7					34.9	3.51
8					29.6	2.54
9					38.2	4.25
10					9.1	1.19
11					[redacted]	[redacted]
12						
13						
14						
Area Under Serum Conc. Day 5 or 6 to Day 1 or 2	61.7	7.72	25.9	8.59	25.1	3.07

*Day 1 was day of first dose of aztreonam. The first serum specimen was obtained prior to the third or fourth dose, which fell on Day 1 or 2 for various patients.

TABLE 1 (cont.)

SERUM CONCENTRATIONS OF AZTREONAM AND SQ 26,992

Add. Pt. No.	7		8		9	
Treatment	AZ	SQ 26,992	AZ	SQ 26,992	AZ	SQ 26,992
Day						
1	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
2	50.7*	3.92*	17.1*	2.29*	31.3*	8.60*
3	21.3	4.43	36.0	2.77	41.3	3.29
4	23.8	2.64	17.4	3.25	45.4	9.37
5	21.5	3.28	19.6	3.27	33.0	10.9
6	27.1*	2.66*	22.5*	3.31*	70.2*	14.1*
7	35.3	4.31	17.1	3.75	71.8	20.2
8	30.6	4.57	15.6	3.40	84.8	25.2
9	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
10						
11						
12						
13						
14						
Area Under Serum Conc. Day 5 or 6 to Day 1 or 2	30.8	3.81	18.5	3.40	58.9	11.3

Mean trough values don't take into account First Dose of loading.

A. Clinical Laboratory Tests

1. Kidney Function Tests

Renal function data are listed in Table 2 for all Addendum patients. Average serum creatinine values for the first 5 days of treatment, and calculated (using nomogram of Siersbaek-Nielsen et al., 1971) values for creatinine clearance are listed in Table 3. All patients had renal insufficiency prior to therapy with aztreonam. There was a trend towards improved renal function during treatment with aztreonam in most patients; however, renal function remained significantly abnormal throughout therapy. No patients showed any apparent deterioration in renal function during aztreonam treatment.

Patient 1 had a calculated creatinine clearance of 66 ml/min, which exceeded the 50 ml/min guideline specified by the protocol, and was therefore analyzed separately.

2. Liver Function Tests

Table 4 shows results for liver function tests for all Addendum patients. Patient 1 had esophageal cancer and was receiving total parenteral nutrition (TPN), but had no specific diagnosis of liver disease, suggesting that the pattern of mildly elevated transaminases (SGOT was 64 to 85 IU/L, SGPT was 49 IU/L) and total bilirubin (1.3 to 2.1 mg/dl) and markedly elevated alkaline phosphatase (360 to 450 U/L) was TPN-induced.

Patient 7 had markedly elevated values of 900 IU/L for SGOT and 1755 IU/L for SGPT, which were judged secondary to hypotension related to the patient's illness.

Patient 9 had hepatic necrosis secondary to hypotension at the time of rupture of an aortic aneurysm approximately 1 month prior to aztreonam therapy, perhaps explaining the initially elevated values of 87 IU/L for SGOT and 52 IU/L for SGPT one day prior to administration of aztreonam. These test values returned to normal during aztreonam therapy, but increased to 76 IU/L for SGOT and 61 IU/L for SGPT on the fifth day after completion of aztreonam treatment. These latter elevations, although mild, were considered by the clinical investigator to be possibly related to aztreonam.

3. Deaths

Patient 7 died 4 days after discontinuation of aztreonam due to hypotension and acidosis, and Patient 9 died 9 days after the end of aztreonam therapy, also due to hypotension and acidosis. Both deaths were unrelated to the administration of aztreonam, in the judgement of the clinical investigator.

VI. ADVERSE REACTIONS

The clinical investigator considered the post-treatment mild elevations in SGOT and SGPT for Patient 9 to be possibly drug related, but not serious.

Appendix I

DOSAGE AND ADMINISTRATION

AZACTAM (aztreonam) For Injection may be administered intravenously or by intramuscular injection. Dosage and route of administration should be determined by susceptibility of the causative organisms, severity and site of infection, and the condition of the patient.

AZACTAM DOSAGE GUIDE
(ADULTS)

TYPE OF INFECTION	DOSE*	FREQUENCY (hours)
Urinary tract infection	500 mg or 1 g	8 or 12
Moderately severe systemic infections	1 g or 2 g	8 or 12
Severe systemic or life-threatening infections	2 g	6 or 8

*Maximum recommended dose is 8 g per day.

The intravenous route is recommended for patients requiring single doses greater than 1 g or those with bacterial septicemia, localized parenchymal abscess (e.g., intra-abdominal abscess), peritonitis or other severe systemic or life-threatening infections. Because of the serious nature of infections due to Pseudomonas aeruginosa, dosage of 2 g every six or eight hours is recommended, at least upon initiation of therapy, in systemic infections caused by this organism.

A single 1 g dose of AZACTAM administered intramuscularly is effective in the treatment of acute uncomplicated urogenital or anorectal gonorrhea and acute uncomplicated cystitis.

The duration of therapy depends on the severity of infection. Generally, AZACTAM should be continued for at least 48 hours after the patient becomes asymptomatic or evidence of bacterial eradication has been obtained. Persistent infections may require treatment for several weeks; some infections such as osteomyelitis may require therapy for four to six weeks. Doses smaller than those indicated should not be used.

For dosage of other antibiotics that may be used concurrently with AZACTAM, consult the manufacturer's professional literature for full prescribing information (see Preparation of Parenteral Solutions, General). ~~AZACTAM may be administered without dosage adjustment.~~

V. Package Insert Labeling Recommendations:

In a meeting with the firm and HFN-815 on 5/22/86, the following package insert labeling modifications/recommendations, etc. were raised by the Division of Biopharmaceutics in regards to the applicant newly revised package insert that was filed 5/13/86 (Appendix B).

Items A-C were discussed in the presence of HFN-815's reviewing medical officer (Dr. Min) but items D and E were not and therefore still may require further discussion with the firm due to possible safety considerations from a clinical perspective.

Recommendation:

A. On page 3 of 17, it was suggested that the second sentence of new paragraph 3 be modified as follows.

> "In patients with impaired renal function, the serum half-life of aztreonam can be prolonged up to about 7 or more hours."

Resolution:

The firm had a concern that since the drug's half-life could continually increase as renal function decreased, trying to put some general estimate half-life in these patients could still be somewhat misleading. Instead, it was suggested by them that the sentence in question be modified to include a reference to the labeling section that is headed as Renal Impairment where dose adjustment is discussed. This was agreed upon.

Recommendation:

B. In the last paragraph of page 3 of 17 it was suggested that the half-life of the open beta-lactam ring hydrolysis product of aztreonam be given (i.e., 1 day or 25 hours).

Resolution:

The firm agreed to do this.

Recommendation:

C. On page 4 of 17 it was suggested that in addition to the mean drug fluid and tissue levels that are provided in the given table, the drug concentration ranges should also be provided. Studies supporting the pericardial and pleural fluid levels plus the tissue concentrations for atrial appendage, endometrium, fallopian tube, fat, gallbladder, kidney, large intestine, liver, lung, myometrium, ovary, skeletal muscle, skin and sternum were requested.

Resolution:

The firm raised a concern about trying to accurately assign concentration ranges when taking into account that a range of different sampling times were sometimes used. They felt this could make the table overly complicated. The firm agreed to submit the requested fluid/tissue level data for verification purposes.

Recommendation:

D. On page 13 of 17 it was suggested that the following sentence be added under OVERDOSAGE following the first sentence.

"A standard 4-hour hemodialysis treatment could remove 27 to 58% of a dose whereas peritoneal dialysis could remove 6 to 12% over 48 hours.

Response:

The firm was reluctant to this suggestion.

E. Summarized were concerns regarding aztreonam dose adjustment in renal impairment as outlined in Section IV of this review. It was explained to the firm that the questions raised were safety issues and the reviewing medical officer should be involved.

NOTE: In the 5/22/86 meeting the firm supplied additional information for Study Protcol #18,554-38, Addendum A. The additional information that was provided includes results for renally impaired patients who were administered aztreonam under multiple dose conditions (see Section III 4b-4d of this review). These results should be brought to the attention of the reviewing medical officer for the purpose of assessing proposed package insert dose adjustments in renally impaired patients.

From the Division of Biopharmaceutics perspective it is recommended that if the proposed package insert dosing recommendations for renally impaired patients are clinically acceptable, then the labelling should be updated to include in the Renal Impairment Section the following, or something similar. "Studies in renally impaired patients have demonstrated that high trough drug levels can occur along with some potential for accumulation of aztreonam's open beta-lactam ring metabolite. In patients with severe renal dysfunction it is therefore prudent to monitor aztreonam and its metabolite before increasing administered doses."

VI. Overall Recommendation:

The Division of Biopharmaceutics (DB) finds NDA 50-580 that was filed 8/27/84 approvable in that it meets the Agency's Bioavailability and Bioequivalence Requirements as cited under 21 CFR 320. Section V of this DB document should be brought to the attention of the reviewing medical officer for resolution of final printed labelling issues if warranted.

6/6/86
John P. Hunt
Pharmacokinetics Evaluation Branch

RD Reviewed by J.P. Skelly, Ph.D.
FT Initialed by J.P. Skelly, Ph.D.

cc: HFN-815, HFN-226(Hunt), HFN-344(Turner), Chron, Drug, and FOI files.

JPH:smj:[redacted]:06-6-86

Table 4

Dose Proportionality Ratio Comparisons using mean AUC 0-24 and mean Cmax Values

Dose Ratio	AUC 0-24		Cmax	
	Bioassay	HPLC	Bioassay	HPLC
$\frac{2000}{500}$ = 4.0	4.1	5.0	4.2	5.7
$\frac{2000}{1000}$ = 2.0	2.0	2.1	1.9	2.0
$\frac{1000}{500}$ = 2.0	2.0	2.4	2.1	2.8

Note: Deviations from apparent dose proportionality using HPLC AUC values

B. Study Protocol #18,544-2 (Pivotal Study)

1. Title: Single-dose intravenous and intramuscular metabolic kinetic study of ^{14}C-aztreonam in healthy subjects.

2. Objective: Define aztreonam's pharmacokinetics, metabolism and excretion following IV and IM doses of ^{14}C-labeled drug.

3. Study Design: Six healthy non-obese male volunteers between the ages of 21 and 30 years (mean weight = 73.9 kg) participated in this study. Each subject received a single 500 mg IV dose and a single 500 mg IM dose of ^{14}C-aztreonam in a randomized crossover study design (3 given IM and 3 given IV per dosing day). Actual doses administered are given in Table 5. There was a 15 day washout period between each drug administration. The IV dose was given as a 2 minute infusion and the IM dose was given in the gluteus maximus.

The drug was supplied as a powder blend of ^{14}C aztreonam and L-arginine containing 50 Ci of radioactivity per 500 mg. The weight ratio of aztreonam to L-arginine in reconstituted solution (sterile water) was 1.0/0.7. Serial serum (0-16 hours), urine (0-144 hours), and fecal samples (0-144 hours) were collected. Serum and urine samples were assayed both by the microbiological method and by radiochemical methods, whereas fecal samples were only assayed by radiochemical methods. Serum protein binding assays were also conducted in this study. Concentrations of total radioactivity were determined by liquid scintallation and concentrations of unchanged drug and its metabolites in serum, urine and feces were determined by thin-layer radio-chromatography.

NOTE: For serum samples for unchanged azetreonam, a correction factor was required (1.0/0.705). "Recovery of intact azetreonam in the serum was in the range of 67 to 75% with an overall mean of 70.5%. An average of about 14% of the radioactivity was recovered as SQ 26,992 (i.e. metabolite) in the same samples that were spiked with ^{14}C-azthreonam regardless of the concentration". The firm indicates, "that some SQ 26,992 (about 14%) was being generated from azthreonam in the actual samples, ..." (NDA Vol. 3.6, pages 2-3689 to 23692; Attachment I).

This study was conducted under the direction of A.A. Sugerman, M.D., Medical Center at Princeton, Princeton, NJ.

4. Results:

a. Table 6 gives mean serum concentrations for total radioactivity, aztreonam (by TLC), SQ 26,992 (major metabolite), and other unknown metabolites following IV administration.

b. Table 7 gives mean serum concentrations for total radioactivity, aztreonam (by TLC), SQ 26,992, and other unknown metabolites (from [redacted] pages [redacted] and [redacted] following IM adminstration. Table 12 gives mean aztreonam concentrations determined by both radio-assay and bioassay for both the IV and IM routes.

and

c. Table 8 gives [illegible]y of the biotransformation profile of aztreonam in urine [illegible]eces.

d. Table 9, 10 and 11 give pharmacokinetic parameters for aztreonam following IV and IM administration.

e. Tables 12 and 13 given serum protein binding results for IV and IM drug administration, respectively.

f. Figures 3 and 4 give mean serum levels of total radioactivity and aztreonam determined by bioassay, respectively. Figure 6 gives mean cummulative urine and fecal excretion of total radioactivity by both the IV and IM routes.

Figure 7 gives mean cummulative renal excretion data for aztreonam and its major metabolite, SQ, 26,992. Figure 8 gives bar charts of mean excretion and biotransformation of IV and IM aztreonam doses. Figure 9 provides mean urinary excretion rate profiles for both the parent drug and to major metabolite.

5. Comments:

a. This study demonstrated that approximately 90% or more of administered ^{14}C-aztreonam (both as IV and IM 500 mg doses) can be accounted for by renal and fecal excretion (Table 8). Over 144 hours about 77% of the administered total radioactivity was recovered in urine and about 13% was recovered in feces. Of that excreted in urine about 67% is parent drug, 7% was metabolite SQ 26,992 and 3% was unidentified metabolites (4 to 5). In feces about 1% was parent drug, 3% SQ 26,996 and 8% as unidentified metabolites.

b. Serum level data for aztreonam as determined by both bioassay and TLRC (radioassay) methods were pharmacokinetically fit by an open 2 compartment model for the IV doses and by an open one compartment model with first order absorption for the IM doses. The pharmacokinetic (PK) parameters generated for this study's IV doses; (Table 9) were in close agreement with the PK values generated for Study Protocol #18,554-1. From serum level data, the mean $t_{1/2}$ values for unchanged aztreonam were about 1.7 hours for both routes of administration.

c. Following IM administration peak drug levels occur about 1 hour post-dose. For the single 500 mg IM dose a mean peak serum concentration of about 20 mcg/ml was achieved. The relative extent of aztreonam absorption following IM administration as compared to IV administration was about 100% (Table 11).

d. Renal clearance for aztreonam was about 70 ml/min (i.e., uncorrected for protein binding). Assuming a normal creatinine clearance value of 120 ml/min, it appears that there may be some net drug tubular reabsorption for aztreonam.

e. Over a concentration range of [redacted] mcg/ml of ^{14}C-aztreonam equivalents; serum binding was determined to be about 70% (Tables 12 and 13).

f. The half-life of the major metabolite, SQ 26,992, is about 25 hours based upon urinary excretion data (Figure 9).

g. Fecal excretion of some parent drug and metabolites following parenteral administration suggests that there be some biliary excretion occurring.

6. Conclusion:

Study #18,554-2 is an acceptable study in that it described the pharmacokinetics, biotransformation and excretion pathways for aztreonam and its metabolites following single IV and IM doses (500 mg). The absolute bioavailability of the IM route was defined in this study (i.e., 100%).