CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER: 21-153/ 21-154

PHARMACOLOGY REVIEW(S)

1Ucl84

NDA 21,153 Review #1

Sponsor & Address: AstraZeneca LP
Wayne, PA

AUG 23 2000

Reviewer: Ke Zhang, Ph.D.
Pharmacologist, HFD-180

Date of Submission: Initial submission: December 3, 1999
Amendment: December 10, 1999
Amendment: April 3, 2000

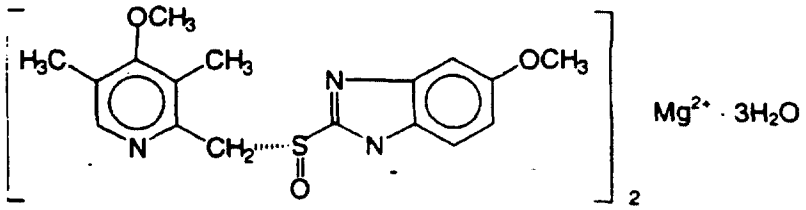
Date of HFD-180 Receipt: Initial submission: December 6, 1999
Amendment: December 13, 1999
Amendment: April 3, 2000

Date of Review: August 8, 2000

REVIEW AND EVALUATION OF PHARMACOLOGY AND TOXICOLOGY DATA
Original Summary

DRUG: Nexium / Esomeprazole magnesium / H 199/18, Delayed-Release Capsules

Bis(5-methoxy-2-[(S)-[(4-methoxy-3,5-dimethyl-2-pyridinyl) methyl]sulfinyl]-1*H*-benzimidazole-1-yl) magnesium trihydrate



Molecular Formula: $C_{34}H_{36}N_6O_6S_2Mg \bullet 3H_2O$ MW: 767.2

CATEGORY: Gastric parietal cell H^+/K^+-ATPase inhibitor.

Related INDs: IND ———

Marketing Indications and Dose: Esomeprazole magnesium is used for treatment of gastroesophageal reflux disease (GERD). For symptom resolution and healing in patients with erosive esophagitis, the recommended dose is — mg once a day for 4-8 weeks. For prevention of the relapse and maintenance of symptom resolution of erosive esophagitis, the recommended dose is — mg

once a day. For treatment of symptomatic GERD, the recommended dose is 20 mg once a day for 4 weeks. The dose of — mg/day represents a dose of — mg/kg/day if 50 kg body weight is assumed.

PRECLINICAL STUDIES AND TESTING LABORATORIES:

Type of Study	Study #	Lot #	Lab	Review Page #
Pharmacology				3-6
Absorption, Distribution, Metabolism And Excretion				6-11
Pharmacokinetic study in rats	4146			
Pharmacokinetic study in rats	4145			
Pharmacokinetic study in dogs	23870			
Excretion and metabolism in dogs	23992			
Acute Toxicity				11-13
Oral toxicity study in rats	T2816	600/93	1	
I.v. toxicity study in rats	T2821	600/93	1	
Subacute/Subchronic Toxicity				
1-month oral toxicity study in Wistar rats	93148	600/93	1	13-19
3 month oral toxicity study in Wistar rats	97477	602/97,102/98	1	19-25
13-week oral toxicity study in Sprague Dawley rats	265/84	HT0916-01-03-01 HT0917-01-03-01 HT0887-01-01-03 HT0888-01-01-02	2	25-32
3-month oral toxicity study in dogs	97103	602/97	1	32-36
Reproductive Toxicity				
Oral embro-fetal development study in rats	97469	602/97	1	36-39
Oral embro-fetal development study in rabbits	98498	1300/98	1	39-45
Mutagenicity				
Ames test	T2817	600/93	1	45-46
In vitro cytogenetic test using human peripheral blood lymphocytes	98045	602/97	3	47-50
In vitro cytogenetic test using human peripheral blood lymphocytes	524/12	600/93	2	50-52
In vivo chromosome aberration test using bone marrow in rats	98457	602/97	2	52-53
Mouse micronucleus test	97484	602/97,102/98	1	53-54

1 = Astra, Sodertalje, Sweden, 2 = — 3 = Astra Charnwood, Leics, England.

Following studies were previously submitted to IND —
(1) pharmacology, (2) ADME, (3) acute toxicity studies in rats, (4) 1-month oral toxicity study in rats, (5) 3-month oral

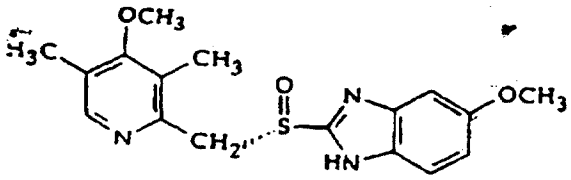
toxicity study in dog, (6) oral dose ranging embryo-fetal development studies in rats and rabbits, (7) Ames test, (8) in vitro chromosome aberration test, and (9) mouse micronucleus test. These studies were previously reviewed on April 2, 1998 (initial submission) and on February 9, 1999 (amendment # 059). These reviews are incorporated in the appropriate portion of the present review.

PHARMACOLOGY:

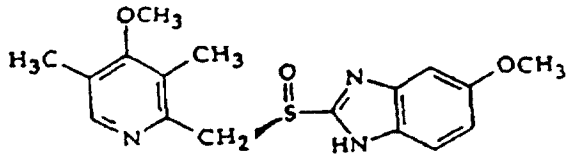
H 199/18 is a S-enantiomer of the racemic proton pump inhibitor, omeprazole. It inhibits H^+/K^+-ATPase activity in gastric parietal cells and thus blocks the final step of the gastric acid secretion. The R-enantiomer is H 199/19. The structures of both enantiomers are shown in the following figure.

Structures of H 199/18 and H 199/19 enantiomers.

H199/18



H199/19



Pharmacological activities of these enantiomers were characterized in *in vitro* and *in vivo* preparations.

Primary Activity

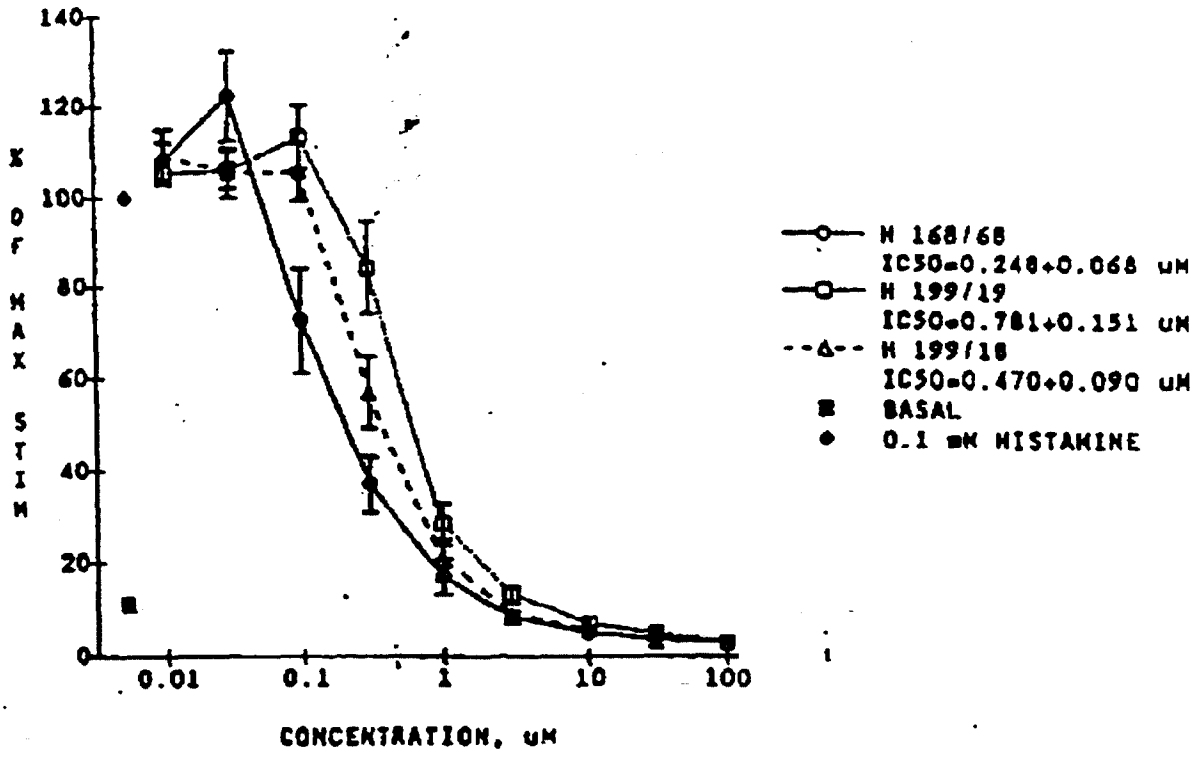
In Vitro Studies:

Effects of Omeprazole and Its Enantiomers on Acid Formation in Isolated Glands:

This study examined the *in vitro* effects of omeprazole and its 2 enantiomers on acid formation in isolated gastric glands of rabbits using an established model (Berglindh et al Acta Physiol Scand 96: 150, 1976). In this model, ^{14}C-aminopyrine (AP), which

is in the form of a weak base in parietal cells (at physiological pH), is protonated under acid secretion and accumulates in gastric glands, which is measured. All 3 compounds significantly inhibited histamine-induced acid formation in a dose-related manner in isolated gastric glands of rabbits (see figure below). The IC_{50} values for H199/19 (0.27 ± 0.052 mg/l) were not significantly different from IC_{50} of H199/18 (0.162 ± 0.031 mg/l). However, significant differences were found between IC_{50} values of omeprazole (0.086 ± 0.023 mg/l) and H199/19. The potency of omeprazole was similar to its S-enantiomer (H199/18). These studies suggest that inhibitory effects of omeprazole on acid secretion can be attributed to both its enantiomers.

. EFFECT OF H 168/68, H 199/19 AND H 199/18 IN GASTRIC GLANDS (n=5).



In Vivo Studies:

Gastric Acid Secretion After a Single Dose of Omeprazole Sodium, H199/18 Sodium, or H199/19 Sodium in Female Rats:

This study compared the in vivo inhibitory effects of 3 drugs (omeprazole sodium, H199/18 sodium and H199/19 sodium) in a rat model. In rats (fasted female Sprague-Dawley) surgically implanted with chronic gastric fistula (in the lumen close to the glandular part of the stomach wall), gastric acid secretion was stimulated by a 2-hr subcutaneous infusion of pentagastrin and carbachol. The acid output (expressed as µmol H^+/30 min) in gastric juice of these rats was calculated from volume (ml/30 min) measured as weight in grams, and sample acidity (mmol/L, determined by titration to pH 7.00 in 30-min fractions). Four groups of rats (n=10/group) were given oral (7 µmol/kg) or iv (1 µmol/kg) dose of either vehicle or the test substance at 75 min before the start of stimulation of the gastric acid secretion. Rats had 1-week of washout period between oral and iv tests. These doses were selected from the dose-range finding studies based on ED_{50} values for acid output of omeprazole sodium. All 3 drugs, i.e. omeprazole sodium, H199/18 sodium, and H199/19 sodium caused a significant inhibition of gastric acid secretion (181, 182 and 133 µmol H^+/30 min resp) (see Table 19 reproduced from volume 1.4, page 96 in the current NDA).

TABLE 19
Gastric Acid Output (Mean ± SEM) After Single Administration of H 199/18, H 199/19 or Omeprazole to Rats

Admin. route	Compound	Dose levels		Gastric acid output
		µmol/kg	mg/kg	µmol H^+/30 min
i.v.	Vehicle	0	0	237±18
	H199/18	1.0	0.35	168±11
	H199/19	1.0	0.35	143±10
	Omeprazole	1.0	0.35	157±17
Oral	Vehicle	0	0	256±19
	H199/18	7.0	2.4	182±15
	H199/19	7.0	2.4	133±22
	Omeprazole	7.0	2.4	181±14

Both oral (181 vs 182 µmol $^+$H/30 min) and iv (157 vs 168 µmol $^+$H/30 min) omeprazole sodium and H199/18 sodium caused a similar acid output, whereas H199/19 sodium was slightly more potent, but no significant differences in antisecretory potency of 3 drugs were noted.

ABSORPTION, DISTRIBUTION, METABOLISM AND EXCRETION (ADME):

RAT:

Study of In Vivo Racemization of H199/18 and H199/19 in the Rat:
(Study # 4146, Report # 3222-0320)

Methods: To examine if there was a partial inversion or racemization of H199/18 or H199/19, rats (Sprague-Dawley, 4/sex) were given H199/18 sodium (batch # H9:1) intraduodenally (id) at a dose of 40 µM/kg (~15 mg/kg). The id route was chosen to avoid the acidic environment of the stomach, and to expose the compounds to the metabolic first-pass effect of the liver. In another group, rats was given H199/19 sodium (batch # H13:2) were given intraduodenally at a dose of 40 µM/kg (~15 mg/kg). Blood

samples were collected (by cannulation of the left carotid artery) at baseline and at 5, 10, and 15 min, and at sacrifice time (20-29 min). Both H199/18 and H199/19 were measured in plasma using ——————————————— method. The percentage of one enantiomer compared with the percentage of the other enantiomer was expressed as the enantiomer excess (EE), which is the difference between the amounts of the S and R-enantiomers or vice versa.

Results: Approximately 2% of the test drugs was determined as the other enantiomer in plasma. In male rats, the EE was 97.4-98.6% for H199/18 or 97.8-99.6% for H199/19. In females, these values were 95.2-98.8% (H199/18) or 97-99.6% (H199/19). These results indicated there is very limited racemization or partial inversion of H199/18 or H199/19 in rats.

Pharmacokinetic Study of Omeprazole Sodium, H199/18 Sodium, and H199/19 Sodium Following Single Intravenous and Intraduodenal Administration in the Rat
(study # 4151, Report # 3222-0336)

Methods: In male Sprague-Dawley rats (n=5), omeprazole sodium (batch # 51), H199/18 sodium (batch # 600/93), and H199/19 sodium (batch # 600/93) were given intraduodenally (id) or intravenously (iv) at a dose of 40 µM/kg (~15 mg/kg). Blood samples were collected (by cannulation of the left carotid artery) at 0, 2, 5, 10, 20, 40, 60, 90, and 120 min after dosing. Both H199/18 and H199/19 were measured in plasma using ——————————————— method (since earlier study showed that no racemization of 2 enantiomers occurs in rats), with limit of quantitation of —— —— µM/l in 0.25 and 0.5 ml of blood respectively.

Results: In male rats, after id administration, all 3 drugs declined in a similar manner with the terminal half life of ~10 min. The maximum plasma concentration of omeprazole sodium, H199/18 sodium, and H199/19 sodium were reached within 5 min, and were 22, 19 and 25 µM/l respectively. The bioavailability (F) of omeprazole sodium, H199/18 sodium, and H199/19 sodium were 39%, 33% and 49% respectively. The bioavailability of H199/19 was significantly higher than the bioavailability of its S-enantiomer (H199/18). After iv, the volume of distribution at steady state (Vd_{ss}) and total blood clearance (Cl) for all 3 compounds (omeprazole sodium, H199/18 sodium, and H199/19 sodium) were

similar (Vd_{ss} = ——— l/kg, CL = ——— ml/min/kg). The results are summarized in the following table.

Comparative pharmacokinetic parameters of omeprazole sodium, H199/18 sodium, and H199/19 sodium, after single id and iv administration in male rats.

Parameters	ID (mean)			IV (mean)		
	Omeprazole	H199/18	H199/19	Omeprazole	H199/18	H199/19
Vss (l/kg)	--	--	--	0.9	0.8	0.7
Cl (ml/min/kg)	--	--	--	75	75	66
AUC (µmol.min/l)	220	168	283	555	545	605
$T_{1/2}$ (min)	10	9	10	12	11	11
T_{max} (min)	3	3	2	--	--	--
C_{max} (µmol/l)	21.5	19.1	24.9	--	--	--
F (%)	39	33	49	--	--	--

F% = The bioavailability was calculated as the AUC ratio of each drug after id administration to that after iv administration. 1 µmol ≈ 0.35 mg

DOG:

Pharmacokinetic Study of Omeprazole, and H199/18 Magnesium, Following Repeated Oral Administration in the Dog
(study # 23870, Report # 1300)

Methods: To compare the pharmacokinetics of the two enantiomers of omaprazole (H199/18 and H199/19) after omeprazole administration, 6 Beagle dogs (females) were given omeprazole (H168/68, batch # 675-01) at 80 µM/kg (~28 mg/kg) by gastric intubation (0.5 ml/kg) for 5 days. Pharmacokinetics of H199/18 was also compared to omeprazole following H199/18 magnesium (batch # 404/95) at oral dose of 80 µM/kg (~29 mg/kg) by gastric intubation. There was a 10 day wash-out period between the administration of H199/18 and omeprazole. At the end of 5 days, blood was collected at 0, 5, 15, 30, 60, 120, 180, 240, 300, and 360 min after dosing of each drug. Both H199/18 and omeprazole were measured in plasma using ——— method. Both enantiomers were also measured by ——— methods. The plasma samples with the highest concentration of H199/18 were further analyzed for conversion or inversion of H199/18 by chiral methods.

Results: In dogs, after oral administration of omeprazole, the peak plasma concentrations of omeprazole, H199/18 and H199/19 were achieved within ~25 min and declined with a mean elimination half life of 30-35 min. Both mean plasma concentration and AUC values of H199/18 were ~1.8 and 2.3 fold higher than those of H199/19, respectively. After H199/18 magnesium and omeprazole administrations, the C_{max} (47.5 µM/l and 43.9 µM/l respectively) and AUC (2973 µM.min/l and 3823 µM.min/l respectively) of both compounds were comparable. The half life of the two drugs were also similar. The concentration of H199/19 enantiomer was determined to be <1.5% after administration of H199/18 magnesium in dogs, suggesting that very little racemization or inversion of H199/18 to H199/19 occurred in dogs. The results are summarized in the following table.

Comparative pharmacokinetic parameters of omeprazole and H199/18 magnesium, after 5 days oral (28 mg/kg) administration in female dogs

Parameters (mean values)	Omeprazole			H199/18 Magnesium
	Omeprazole	H199/18	H199/19	H199/18
AUC (µmol.min/l)	1925	1329	569	2973
$T_{1/2}$ (min)	37	35	28	32
T_{max} (min)	25.8	25.8	25.8	18
C_{max} (µmol/l)	22.0	14.2	7.7	47.5

AUC was calculated using the log-linear trapezoidal method, from zero to 4-6 hrs. and extrapolated to infinity from the area calculated from elimination constant and the plasma concentration in the latest plasma sample. 1 µmol ≈ 0.35 mg

Excretion and Metabolism of H 199/18-^{14}C in Dogs after Oral Administration - A Comparison With [^{14}C]-Omeprazole (23992)

Methods: To study excretion of H 199/18 and to identify the metabolites, three dogs were given a single oral dose (gavage) of H 199/18-^{14}C at 16 µmol/kg (5.5 mg/kg). One dog received [^{14}C]-omeprazole at 16 µmol/kg (5.5 mg/kg). The special activity of H 199/18-^{14}C was 0.63 kBq/nmol and 1.78 kBq/nmol of [^{14}C]-omeprazole. Urine and faces were collected for up to 120 hours after dosing. The radioactivity was determined using _____ Parent compound and metabolites were identified using _____

Results: The results indicated that 92% (H 199/18) and 87%

(omeprazole) of the radioactivity were recovered within 120 hours after dosing. Of the total recovered radioactivity 61% was in feces and 31% in urine for H 199/18. For omeprazole, 57% of the total radioactivity was in feces and 30% in urine. Both H 199/18 and omeprazole were extensively metabolized. Six major metabolites (glucuronic acid conjugates) were identified in urine which accounted for 54% (H 199/18) or 48% (omeprazole) of the radioactivity recovered in urine during the first 10 hours following dosing. The aglycones of these conjugates were the major metabolites identified in feces which accounted for 26% (H 199/18 and omeprazole) of the radioactivity recovered in feces. The parent compound was accounted for only <1% of the total radioactivity recovered.

To compare the similarities and differences between rats, dogs, and humans, the results of pharmacokinetic studies of H 199/18 and omeprazole are summarized in the following table.

	Rat	Dog	Human
H199/18			
Dose	ID, 15 mg/kg	GI, 29 mg/kg	Oral, 20 mg
ID bioavailability,%	33	---	---
T_{max}, min	3	18	96
C_{max}, mg/l	6.7	16.6	0.74
AUC, mg.min/l	58.8	1040.5	88.2
Terminal $T_{1/2}$, min	9	32	72
Cl, ml/min/kg	75*	---	---
Vss, l/kg	0.8*	---	---
Omeprazole			
Dosing, mg/kg	ID, 15 mg/kg	GI, 29 mg/kg	Oral, 20 mg
ID bioavailability,%	39	---	---
T_{max}, min	3	25.8	78
C_{max}, mg/l	7.5	7.7	0.49
AUC, mg.min/l	77	673.8	48.3
Terminal $T_{1/2}$, min	10	37	60
Cl, ml/min/kg	75*	---	---
Vss, l/kg	0.9*	---	---

ID = intraduodenal, GI = gastric intubation, * = following i.v. dose of 14 mg/kg, the human dose was given to GERD patients, 1 µmol ≈ 0.35 mg.

In summary, the maximum plasma concentrations of H199/18 and omeprazole were reached quickly within 3 minutes after intraduodenal administration in rats and within 18-26 minutes after gastric intubation in dogs. The intraduodenal bioavailability of H199/18 and omeprazole were 33% and 39%, respectively, in rats. The plasma levels of H199/18 and omeprazole declined quickly with terminal half life of 9-10 minutes in rats and 32-37 minutes in dogs. In humans, the plasma

levels of H199/18 and omeprazole declined slower than those in rats and dogs with half lives of 1.1-1.2 hours in GERD patients. In both rats and dogs, very little (1.5-2%) racemization or inversion of H199/18 to H199/19 or vice versa occurs, suggesting that these enantiomers are resistant to inversion. Both H199/18 and omeprazole were excreted mainly in feces (57-61%) and in urine (30-31%).

TOXICITY:

ACUTE TOXICITY:

The Comparisons of Acute Oral and Intravenous Toxicity Of H199/18 Sodium, H199/19 Sodium and Omeprazole Sodium in Rats
(Study # T2816 and T2821)

Methods: The acute toxicity of H199/18 sodium (batch # 600/93), H199/19 sodium (batch # 600/93) and omeprazole sodium (batch # 51) after oral (gavage) and iv administrations were examined in male and female Wistar rats. All 3 compounds were dissolved in distilled water (at concentration of 70, 140 and 270 µM/ml, pH 10.6-11.3) and saline (pH 10.7-10.9, and given in volumes of 20 ml/kg) for oral and iv administrations, respectively. Additional 2 groups of rats (n=5/sex) were given omeprazole sodium (310 mg/kg) dissolved in buffered polyethylene glycol 400 (PEG400, pH 10.3) or PEG400 alone (control animals) by iv administration. The dosing information is summarized in a table in the result section. All animals were observed for toxic signs and mortality daily for 14 days. At the end of observation period, all animals were necropsied and subjected to standard examinations. In addition esophagus, stomach, duodenum, jejunum, ileum, cecum, colon and rectum were examined microscopically.

Results: After oral administration of H199/18, deaths were noted at 510 (2 females died), 990 (one male + 3 females), and 2000 (4 males died) mg/kg. Deaths with H199/19 were found at 990 (1 M + 1 F died) and 2000 (4 M + 5 F died) mg/kg. Deaths after omeprazol were 1 male and 5 females at 990 mg/kg and 3 males at 2000 mg/kg. All 3 drugs (at all oral doses) caused similar clinical signs (reduced activity to unconsciousness, coupled with reduced respiratory frequency and abdominal respiration). Also, repeated clonic convulsions (associated with tremors, salivation, dyspnea, cyanosis, ataxia and/or reduced motor activity). were observed at all doses of these 3 compounds, except at 510 mg/kg

of H199/19 sodium. Convulsions (which lasted for 5 seconds) were usually first observed within few min and continued intermittently for up to 1-3 hrs. Females in general had clinical signs (also had lacrimation and dark colored urine) with greater intensity and duration, as well as increased mortality ratios. All surviving animals recovered from these abnormalities within 23 hrs after treatment. Since no macroscopic changes were noted, no histopathology in GI tracts were examined.

After iv administration, all 3 drugs (H199/18 sodium, H199/19 sodium and omeprazole sodium) showed similar clinical signs (at all doses, including piloerection and lacrimation at higher doses) as noted after oral administration. A marked local reaction at the injection site was observed in the tail in all drug treated animals and incidence and severity were increased with the increase in dose and pH. In many animals it developed into ulcerations. With omeprazole sodium given in PEG400, a more pronounced reaction was observed with death within 4 min (all 10 rats died, vs none with the vehicle PEG400 alone). The vehicle PEG400 itself caused reduced motor activity and increased respiratory frequency in rats. Some animals displayed pulmonary congestion due to agonal circulatory insufficiency. The mortality rate is summarized in the following table along with the dosing information.

Mortality rate in oral dose study in rats

Species (strain) Wistar Rats	No/Sex/Group	Doses (mg/kg)	Mortality rate	Minimum Lethal Doses (mg/kg)
H199/18 Sodium				
Males	5	510 990 2000	0 1 4	990
Females	5	260 510 990	0 2 3	510
H199/19 Sodium				
Males & Females	5	510 990 2000	0 1 male, 1 female 4 males, 5 females	990
Omeprazole Sodium				
Males & Females	5	510 990 2000 (males)	0 1 male, 5 females 3 males,	990

Mortality rate in the intravenous dose study in rats

Species (strain) Wistar Rats	No/Sex/Group	Doses (mg/kg)	Mortality rate	Minimum Lethal Doses (mg/kg)
H199/18 Sodium				
Males & Females	5	180 310	0 1 male, 2 females	310
H199/19 Sodium				
Males & Females	5	180 310	0 5 males, 1 female	310
Omeprazole Sodium				
Males & Females in saline	5	180 240 310	0 1 male 2 males, 2 females	240 for males 310 for females
In PEG400		310	5 males, 5 females	

In conclusion, the minimum lethal oral doses were 990 mg/kg for all three drugs except for the females following H199/18 sodium (510 mg/kg). The minimum lethal iv doses were 310 mg/kg for all three drugs except for the males after omeprazole (240 mg/kg). Major clinical signs were reduced motor activity, increased or decreased respiratory frequency, abdominal respiration, clonic convulsions in association with tremors, salivation, dyspnea, cyanosis, and ataxia observed after both oral and iv doses. Clinical signs in females in general were of greater intensity and duration. All surviving animals recovered from these effects within 22-23 hrs after treatment.

SUBACUTE/SUBCHRONIC/CHRONIC TOXICITY:

The Comparative 1-Month Oral Toxicity Studies of H199/18 Sodium, H199/19 Sodium, Omeprazole Sodium, and Omeprazole in Rats
(Study # 93148, report # T2823)

Testing Laboratories: Astra Hassle AB,
Sodertalje, Sweden.

Study Started: December 14, 1993

Study Completed: June 2, 1994

GLP Requirement: A statement of compliance with the GLP regulations and quality assurance unit was included.

Animals: Wistar rats (~7-8 weeks old), males 180-270 g, females 130-210 g.

Drug Batch No.: H199/18 sodium, batch # 600/93,
H199/19 sodium, batch # 600/93,
Omeprazole sodium, batch # 51.
Omeprazole, batch # 599 and 660

Methods: The aim of this study was to compare the oral 1-month toxicity of H199/18 sodium (the s-enantiomer of omeprazole) with that of H199/19 sodium (the R-enantiomer). In addition, the effects of the above 2 compounds were compared with omeprazole sodium and omeprazole (neutral form) in rats. Four groups (groups 1-4) of 10 male and 10 female rats were given oral H199/18 sodium by gavage at 0, 15, 48, and 150 mg/kg/day (0, 40, 130, and 400 µM/kg) for 1 month at volumes of 10 ml/kg body weight/day. Three groups (groups 5-7) of 10 males and 10 females received the same dose of H199/19 sodium. Two additional groups (groups 8-9) of 10 males and 10 females received omeprazole sodium or omeprazole (batch # 599 for first 6 days, and batch # 660 during the remainder period of the study) at 150 mg/kg/day. H199/18 sodium, H199/19 sodium, and omeprazole sodium were dissolved in purified water at concentration of 4, 13 and 40 µM/ml, whereas omeprazole was formulated as 40 µM/ml suspension in 0.5% carbonate buffered hydroxypropyl methyl cellulose solution. Control groups received the tap water only. The dosing solutions from the first and the last day were analyzed for concentration, pH, stability and chiral purity of the test compound. The concentrations of the test substances were 93-99% of the intended doses, pH varied between 8.3-9.1, and all compounds were stable throughout the study. Additional 9 groups of satellite animals (groups 10-18, n=6/dose/sex) were used for toxicokinetics and evaluation of thyroid hormone levels (study # 93170, report # T2822). These rats similarly received the same doses of all the drugs for 1-month. Mortality and clinical signs were observed once daily. Body weights and food consumption were noted once weekly, and water consumption once during weeks 1 and 3. Ophthalmological examinations were carried out on all rats before and at the end of the study. Eyes were examined with an indirect ophthalmoscope after application of 0.5% solution of tropic amide. Hematology, clinical chemistry

tests, and urinalysis (following deprivation of food) were carried out after about 3 weeks. Gross pathology and complete histopathological examinations were carried out on all animals. For toxicokinetics, blood was collected from the orbital venous plexus of the 4 male and 4 female satellite rats (groups 10-18) at 10, 20 and 60 min after dosing on day 0, and of 4 males and 4 females of groups 1-9 at 10, 20 and 60 min after dosing following 1-month of treatment (on days 32-35). The plasma concentration of omeprazole, H199/18, and H199/19 were measured by [redacted] method with limit of quantification of [redacted] µmol/l of plasma. Blood for thyroid hormone levels were collected from 6 male and 6 female rats in each of 10-18 groups on days 25 and 26 of dosing. Plasma levels of triiodothyronine (T3), thyroxine (T4) and thyroid stimulating hormone (TSH) were determined by established methods.

Results:

1. Observed Effects: At 150 mg/kg/day, all 4 compounds caused increased salivation in few rats during the last 2 weeks of the study. This was observed shortly before or an hour after dosing.

2. Mortality: One rat died in the group of 15 mg/kg/day of H199/18 on day 6. Two rats died in the group of 48 mg/kg/day of H199/19 on days 4 and 6. Two animals died due to bronchopneumonia and 1 due to circulatory dysfunction (which had pulmonary congestion). None of these were treatment related.

3. Body Weight/Food Consumption/Water Consumption: The initial and final (week 4) mean body weights were 208.2 g and 313.4 g (male) or 165.3 g and 211.6 g (female), respectively in the control group. In males, at 48 mg/kg/day of H199/18 and H 199/19, there was a significant decrease in the body weight gains (with H199/18 by 14.5%, with H199/19 by 8.6%). At 150 mg/kg/day, all 4 compounds caused a significant decrease in the body weight gains in both males (by 24-25.9% and 24% with H199/18 and H199/19, 12.7-16.3% with omeprazole sodium and omeprazole) and females (by 19.6-25.2%).

The initial and final mean food consumptions were 22.8 g/animal/day and 25.7 g/animal/day (male) or 18 g/animal/day and 19.8 g/animal/day (female), respectively. At 150 mg/kg/ day, food consumption was lower in females with all 4 compounds (by

27.8-44.4%). The individual variability in water concentration was large, and therefore no conclusion could be drawn.

4. Ophthalmoscopy: No treatment related effects were observed.

5. Hematology: At 150 mg/kg/day, a slight increase in mean blood platelet count was observed in males (by 9-17%) and in females (by 18-24%) with all 4 compounds.

6. Blood Chemistry/Urinalysis: No treatment related changes in serum or urinalysis were observed.

7. Organ Weights: Slight increase in the stomach (11-18%) and thyroid weights (16-42%) were in all 4 drug groups mainly at high dose. Slight decrease in the spleen, thymus, and ovary weight were also noted at high dose for all four compounds.

8 Gross Pathology: No treatment related effects were observed.

9. Histopathology: At high dose with all 4 compounds, minimal foci of chief cell eosinophilia in gastric mucosa were observed in 1-2 males and/or females (H199/18 sodium 1 of 10 males, H199/19 sodium 1 of 10 males, omeprazole sodium 2 of 10 males, and omeprazole 1 of 10 females).

10. Toxicokinetics: The peak plasma concentrations were observed at 10 min after dosing. The $AUC_{0\text{-}60min}$ values of H199/18 and H199/19 were higher in females (by ~4-12 times) than in males, and increased in both sexes unproportionately with increases in doses (by 3-7 fold) at 1-month. AUC values were highly variable (M±SD) with all 4 compounds. At 150 mg/kg/day, on day 0, no significant differences in AUC of 4 compounds were observed. However, after 1-month, the AUC values at this dose were significantly higher in males with omeprazole sodium than with H199/18 sodium (1690 vs 172 μmol.min/l), which were due to a large variability in individual AUC caused by 1 of 4 males with omeprazole sodium. The results are summarized in the following table.

Mean plasma C_{max} and AUC values of H199/18, H199/19 (15, 48 and 150 mg/kg/day), omeprazole sodium and omeprazole (150 mg/ kg/day) following oral (by gavage) administration (1 µmol ≈ 0.35 mg)

Day 0	Sex (M/F)	15 mg/kg/day	48 mg/kg/day	150 mg/kg/day
H199/18 Sodium C_{max} (µmol/l)	M	0.30	3.18	16.7
	F	3.40	11.70	65.8
AUC Mean ± SD (µmol.min/l)	M	4.54	51.2	313 ± 168
	F	52.60	221.0	2370 ± 1340
H199/19 Sodium C_{max} (µmol/l)	M	1.66	3.40	25.4
	F	9.01	11.40	58.6
AUC Mean ± SD (µmol.min/l)	M	25.1	69.2	537 ± 205
	F	167.0	275.0	2060 ± 831
Omeprazole Sodium C_{max} (µmol/l)	M	--	--	25.7
	F	--	--	51.5
AUC Mean ± SD (µmol.min/l)	M	--	--	536 ± 345
	F	--	--	1810 ± 1420
Omeprazole C_{max} (µmol/l)	M	--	--	17.1
	F	--	--	47.7
AUC Mean ± SD (µmol.min/l)	M	--	--	528 ± 280
	F	--	--	2060 ± 680
Day 30	**Sex**	**15 mg/kg/day**	**48 mg/kg/day**	**150 mg/kg/day**
H199/18 Sodium C_{max} (µmol/l)	M	0.39	2.55	7.75
	F	0.86	5.24	27.10
AUC Mean ± SD (µmol.min/l)	M	7.2	51.5	172 ± 55
	F	14.6	107.0	600 ± 280
H199/19 Sodium C_{max} (µmol/l)	M	1.51	5.86	22.5
	F	4.16	18.00	51.4
AUC Mean ± SD (µmol.min/l)	M	25.5	105	636 ± 505
	F	77.8	496	1550 ± 464
Omeprazole Sodium C_{max} (µmol/l)	M	--	--	65.0
	F	--	--	50.5
AUC Mean ± SD (µmol.min/l)	M	--	--	1690 ± 2180
	F	--	--	1460 ± 1000
Omeprazole C_{max} (µmol/l)	M	--	--	10.0
	F	--	--	26.1

AUC Mean ± SD (µmol.min/l)	M	--	--	299 ± 106
	F	--	--	1210 ± 555

11. Thyroid Hormone Levels: No treatment related changes in plasma levels of T3, T4 or TSH were observed as shown in the following table

Mean plasma levels and standard deviations for T3, T4 and TSH in rats.

Group No.	Test compound	Daily dose µmol/kg	Sex	n	Mean levels ± SD T3 nmol/l	T4 nmol/l	TSH ng/ml
1	Vehicle	0	M	6	0.7±0.1	61±8	7.8±1.8
"	"	"	F	6	1.0±0.1	40±10	6.4±0.6
2	H 199/18 sodium	40	M	6	0.8±0.1	61±5	8.0±1.0
"	"	"	F	6	1.0±0.2	33±5	6.8±2.1
3	"	130	M	6	0.8±0.1	60±7	9.1±1.7
"	"	"	F	5	1.0±0.2	39±9	6.8±1.1
4	"	400	M	6	0.9±0.1*	63±7	9.3±1.1
"	"	"	F	6	1.3±0.3*	47±10	6.6±0.8
5	H 199/19 sodium	40	M	6	1.0±0.1*	64±17	9.1±4.1
"	"	"	F	5	1.1±0.2	37±10	6.2±1.5
6	"	130	M	6	1.1±0.2*	65±6	9.9±2.8
"	"	"	F	6	1.2±0.1*	41±3	6.7±0.6
7	"	400	M	6	0.9±0.2*	58±4	9.2±3.4
"	"	"	F	6	1.2±0.2*	37±6	6.4±1.4
8	Omeprazole sodium	400	M	6	1.1±0.1*	76±14*	9.9±2.0
"	"	"	F	6	1.2±0.2*	41±11	6.6±1.1
9	Omeprazole	400	M	6	1.1±0.2*	73±14	8.3±1.8
"	"	"	F	6	1.2±0.2*	34±11	6.2±0.9

* significantly different from corresponding control rats in group 1.

These results indicate that oral administration of H199/18 sodium (15, 48 and 150 mg/kg/day), H199/19 sodium (15, 48 and 150 mg/kg/day), omeprazole sodium (150 mg/kg/day) and omeprazole (150 mg/kg/day) for 1-month caused a significant decrease in the body weight gains (males by ~13-26%, females by 20-25%), clinical signs (increased salivation), and histopathological chagnes in

the stomachs (minimal foci of the chief cell eosinophilia in gastric mucosa at 150 mg/kg/day doses of all 4 compounds). The effects of all 4 compounds at 150 mg/kg/day were similar. Also at 48 mg/kg/day, both H199/18 sodium (by 14.5%) and H199/19 sodium (by 8.6%) caused a significant decrease in the body weight gains in males. The `no effect doses' of H199/18 and H199/19 were 15 mg/kg/day in males and 48 mg/kg/day females.

3-Months Oral Toxicity Study of H 199/18 in Wistar Rats
(97477)

Testing Laboratories: AstraZeneca R & D,
Södertälje, Sweden

Study Start and Completion Dates: February 5, 1999 and -
July 6, 1999

GLP Requirement: Sponsor included a statement of compliance with GLP regulations and a quality assurance statement.

Animals: Male (180-290 g, ~8 weeks old)
Female (140-200 g, ~8 weeks old)
Wistar rats

Drug Batch No.: 602/97, 102/98

Methods: To evaluate the toxicity of H 199/18 in rats, H 199/18 was given by oral gavage to rats (10/sex/group) at 0, 40, 200, and 800 µmol/kg/day (14, 69, and 280 mg/kg/day) for three months. A separate group received ompeprazole by oral gavage at 400 µmol/kg/day (140 mg/kg/day). All animals were observed daily. Individual body weights and food consumption were recorded weekly. Water consumption was measured during weeks 0, 3, and 8. Ophthalmoscopy was performed before dosing started and at the end of the study. Hematology, blood chemistry and urinalysis were performed after about 1 and 3 months of dosing. All animals were necropsied on the day following the last day of dosing. The organs were weighed at necropsy. Histopathological examination was conducted in the control, high dose, and omeprazole groups. Histopathological examination was also conducted in the prematurely dead or sacrificed animals and the animals with relevant gross changes. The stomach and kidneys were microscopically examined in all animals. In the separate satellite groups, plasma concentrations of H 199/18 were determined on day 1 and at termination.

Results:

1. Clinical signs of toxicity: Ploughing the nose through the cage bedding and salivation were noted shortly after dosing in the mid and high dose groups.

2. Mortality: One female in the mid dose group died right after dosing. Two females (one control and one high dose) were sacrificed due to eye damage after blood sampling. These deaths were not considered as treatment related.

3. Body Weight: The initial and final body weights were 210 and 392 g in control males or 161 and 230 g in control females. Terminal body weight gain was lower mainly in the treated males (16%, 13%, 28%, and 18% in the low, mid, high, and omeprazole groups, respectively) than that in the control. Slightly decreased terminal body weight gain was also noted in the high dose females (7%) as compared to the control.

4. Food Consumption: Food consumption in the control group was 20-23.3 g/animal/day in males or 14.1-16.9 g/animal/day in females. Slight decreased food consumption was noted in the high dose group.

5. Water Consumption: No clear treatment related changes were noted.

6. Ophthalomoscopy: There were no treatment related changes.

7. Hematology: The mean concentrations of hemoglobin (Hgb), hematocrit (Hct), mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH) were slightly decreased in the treatment groups as compared to the control. Mean platelet count was higher in the treatment groups as compared to the control. These results were summarized in Table 7 on page 42 in volume 16. This table is attached below.

Table 7 Mean haematological values after 4 and 12 weeks' treatment

Measurement code (unit)	Week	Males					Females				
		1	2	3	4	5	1	2	3	4	5
B-Hgb (g/L)	4	157	158	152*	148**	150*	152	149	148	143**	143**
	12	147	150	139**	131**	132**	139	139	132	126**	125*
B-Hct (x1)	4	0.46	0.46	0.45	0.44*	0.44*	0.44	0.44	0.44	0.43	0.43
	12	0.44	0.45	0.42*	0.41*	0.41*	0.42	0.42	0.40	0.39*	0.39*
B-MCV (fl)	4	53.9	54.4	51.1*	51.6*	51.0*	55.6	55.2	55.3	54.9	55.4
	12	53.4	53.8	50.8*	48.7*	49.8*	57.9	56.6	53.5*	50.9**	54.0**
B-MCH (pg)	4	18.6	18.7	17.5**	17.6**	17.6**	19.3	18.9	18.7*	18.3**	18.6*
	12	18.1	18.1	16.9**	15.9**	16.7**	19.4	18.8	17.6**	16.4**	17.7**
Platelets (x10E9/L)	4	947	998	1058*	1045	1067	974	1035	1077	1048	1023
	12	907	920	1016	1120**	1011*	913	1005	1126**	1134**	1067*

* $p < 0.05$ ** $p < 0.01$

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

8. Clinical Chemistry: Serum glucose was decreased (20-22%) in males treated with H199/18 (high dose) and omeprazole at week 4 after dosing as compared to the control. This was also seen at week 12 (16-17%). Slight decrease of serum glucose (15%) was also noted in the high dose females of H199/18 at week 12. Slight increase in serum cholesterol was seen in the high dose group of H199/18 as compared to the control Serum bilirubin was increased in the high dose group of H199/18 as compared to the control Total serum protein was also increased in the treated females as compared to the control

Serum gastrin level was increased in treatment groups and these results were summarized in Table 9 on page 44 in volume 16. This table is attached below.

Table 9 Mean serum gastrin concentrations (2 h after dose) ±SD after 12 weeks' treatment

	Males					Females				
	1	2	3	4	5	1	2	3	4	5
Gastrin (pg/ml)	90 ±22	340 ±190	1100 ±400	2000 ±400	1300 ±200	67 ±28	500 ±250	1200 ±200	2200 ±400	2000 ±300

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

9. Urinalysis: Slightly higher urinary osmlarity was noted in the high dose and omeprazole females (497-524 mOsm/kg, control = 370 mOs/kg).

10. Organ Weights: The absolute stomach weight was increased by 8.4%, 25.3%, 25.8%, and 22.6% (male) or 16.5%, 26.3%, 48%, and 30% (female) in the low, mid, high dose groups of H 199/18, and omeprazole group as compared to the control, respectively (absolute stomach weight = 1.9 g in control males or 1.52 g in control females). Increased absolute liver weight (25.7%) was found in the high dose females as compared to the control (absolute liver weight = 11.37 g in control males or 7.61 g in control females). Decreased absolute thymus weight was noted in the high dose males (37.5%) as compared to the control (0.33 g). The absolute spleen weight was decreased by 10.4%, 12.6%, 23%, and 21% in the low, mid, high dose groups of H 199/18, and omeprazole group as compared to the control, respectively (absolute spleen weight = 0.74 g in control males or 0.66 g in control females).

11. Gross Pathology: No gross changes were observed.

12. Histopathology: Histopathological examination revealed treatment related changes in the stomach and kidney and these results were summarized in Table 11 on page 47 in volume 16. This table is attached below.

Table 11 Incidence (no. of animals affected) of treatment-related histopathological findings in the stomach

Sex		Males					Females				
Group		1	2	3	4	5	1	2	3	4	5
No. of animals		10	10	10	10	10	10	10	10	10	10
STOMACH											
Chief cell eosinophilia	Total	-	-	5	10	9	-	-	2	10	10
	Minimal	-	-	5	2	6	-	-	2	2	10
	Slight	-	-	-	4	3	-	-	-	5	-
	Moderate	-	-	-	4	-	-	-	-	3	-
Mean score/group and sex		0	0	0.5	2.2	1.2	0	0	0.2	2.1	1.0
Vacuolated glandular cells	Total	-	-	-	6	1	-	-	-	6	-
	Minimal	-	-	-	-	1	-	-	-	-	-
	Slight	-	-	-	6	-	-	-	-	6	-
Mean score/group and sex		0	0	0	1.2	0.1	0	0	0	1.2	0

Table 12 Incidence (no. of animals affected) of treatment-related histopathological findings in the kidneys

Sex		Males					Females				
Group		1	2	3	4	5	1	2	3	4	5
No. of animals		10	10	10	10	10	10	10	10	10	10
KIDNEYS											
Basophilic cortical tubules	Total	2	-	2	9	6	-	1	-	7	2
	Minimal	2	-	2	6	6	-	1	-	3	2
	Slight	-	-	-	2	-	-	-	-	3	-
	Moderate	-	-	-	1	-	-	-	-	1	-
Mean score/group and sex		0.2	0	0.2	1.3	0.6	0	0.1	0	1.2	0.2
Inflammatory cell infiltration	Total	1	2	-	3	1	-	-	-	7	1
	Minimal	1	2	-	3	1	-	-	-	7	1
Mean score/group and sex		0.1	0.2	0	0.3	0.1	0	0	0	0.7	0.1

The mean scores/group and sex have been calculated to illustrate the comparative degree of each finding in the different groups. This is the total sum of the various degrees of change (minimal = 1, slight = 2, moderate = 3) times the number of animals affected at each does level and sex, in relation to the number of animals examined at each dose level and sex (10M + 10F).

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

13. Toxicokinetics: Plasma levels of H 199/18 in males were higher after 3 month treatment than those after the first treatment. This difference was not obvious in females. Plasma levels of omperazole in both males and females were higher after 3 month treatment than those after the first treatment. These

results were summarized in Table 4 on page 38 and 39 in volume 16. This table is attached below (1 µmol ≈ 0.35 mg).

Table 4 Dose levels and exposure to H199/18 or omeprazole

Compound	Dose		Median (range) Cmax (µmol/l)		Median (range) AUC(0-2 h) (µmol·min/l)	
	µmol/kg	Mg/kg	Day 0	3 months	Day 0	3 months
Males						
H 199/18	40	14	0.336	0.921	NC	25.4
H 199/18	200	69	3.53	10.6	97.5	273
H 199/18	800	280	14.7	40.7	557	1750
Omeprazole	400	140	14.2	17.9	472	986

NC = Not calculated

Table 4 Dose levels and exposure to H199/18 or omeprazole (continued)

Compound	Dose		Median (range) Cmax (µmol/l)		Median (range) AUC(0-2 h) (µmol·min/l)	
	µmol/kg	Mg/kg	Day 0	3 months	Day 0	3 months
Females						
H 199/18	40	14	0.453	0.683	NC	19.8
H 199/18	200	69	22.9	6.46	473	280
H 199/18	800	280	98.8	122	8160	8450
Omeprazole	400	140	38.7	76.6	2810	5870

NC = Not calculated

In summary, H 199/18 was tested orally in Wistar rats at 0, 14, 69, and 280 mg/kg/day for three months. Major treatment related changes were decreased terminal body weight gain by 16%, 13%, and 28% in the low, mid, and high dose males, respectively. Slight decrease in the mean concentrations of hemoglobin, hematocrit, mean corpuscular volume, and mean corpuscular hemoglobin and increase in the platelet count (up to 24% in high dose males) were noted in the treatment groups. Histopathological changes were mainly in the high dose group and these changes included chief cell eosinophillia and vacuolated glandular cells

in stomach and basophilic cortical tubules and inflammatory cell infiltration in kidney.

13-week Oral Toxicity Study of H 199/18 in Sprague-dawley Rats
(265/84)

Testing Laboratories: [ ]

Study Start and Completion Dates: October 28, 1998 and July 27, 1999

GLP Requirement: Sponsor included a statement of compliance with GLP regulations and a quality assurance statement.

Animals: Male (150-210 g, ~6-7 weeks old)
Female (120-170 g, ~6-7 weeks old)
Sprague-Dawley rats

Drug Batch No.: HT0916-01-03-01, HT0917-01-03-01, HT0887-01-01-03, HT0888-01-01-02

Methods: To evaluate the toxicity of H 199/18 in rats, H 199/18 was given by oral gavage to rats (10/sex/group) at 0, 40, 200, and 800 µmol/kg/day (14, 69, and 280 mg/kg/day) for three months. A separate group received omeprazole by oral gavage at 400 µmol/kg/day (140 mg/kg/day). All animals were observed daily. Body weight, food consumption, and water consumption were recorded weekly. Ophthalmoscopy was performed before dosing started and during week 12. Hematology, clinical chemistry, and urinalysis were performed during weeks 4 and 13. All animals were necropsied at termination. The organs were weighed at necropsy. Histopathological examination was conducted in the control, high dose, and omeprazole groups. Histopathological examination was also conducted in the animals with relevant gross changes. The stomach, liver, spleen, kidneys, and thymus were microscopically examined in all animals in all groups. In the separate satellite groups, plasma concentrations of H 199/18 were determined on day 1 and at termination.

Results:

1. Clinical signs of toxicity: There were no treatment related clinical signs of toxicity observed.

2. Mortality: There were no deaths in this study.

3. Body Weight: The initial and final body weights were 191.8 and 453.5 g in control males or 155.1 and 276.1 g in control females. The terminal body weight gain was comparable in all groups.

4. Food Consumption: Food consumption in control group was 23-30 g/animal/day in males or 18-21 g/animal/day in females. Slight increased food consumption was noted in high dose females and omeprazole group.

5. Water Consumption: Water consumption in control group was 28-36 g/animal/day in males or 22-30 g/animal/day in females. There was dose dependent increase in the water consumption. This was more obvious in the high dose group and omeprazole group.

6. Ophthalomoscopy: There were no treatment related changes.

7. Hematology: Decreases in the mean concentrations of hemoglobin (Hgb), packed cell volume (PCV), mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH) and increase in the mean platelet count were found in the high dose group and omeprazole group as compared to the control. These results were summarized in Table 4 on page 54 in volume 1.17. This table is attached below.

Haematology Table 4, Appendix 4

Measurement code (unit)	Week	Males				
		1M	2M	3M	4M (approximate % relative to control)	5M (approximate % relative to control)
Hb (g/dL)	4	15.8	15.2	15.3	14.0*** (11% less)	14.0*** (11% less)
	13	16.2	15.9	16.0	14.2*** (12% less)	14.6*** (10% less)
PCV (%)	4	48.0	46.3	47.1	43.7*** (9% less)	43.7*** (9% less)
	13	47.8	46.7	46.3	41.5*** (13% less)	42.7*** (11% less)
MCV (fL)	4	58.8	59.3	57.9	54.8*** (7% less)	55.0*** (6% less)
	13	53.5	53.9	51.1	49.0** (8% less)	46.9*** (12% less)
MCH (pg)	4	19.4	19.5	18.8	17.5*** (10% less)	17.6*** (9% less)
	13	18.1	18.3	17.7	16.7* (8% less)	16.0*** (12% less)
Platelets (1000/cmm)	13	931	1084	971	1240** (33% more)	1251** (34% more)

* P<0.05
** P<0.01
*** P<0.001

Measurement code (unit)	Week	Females				
		1F	2F	3F	4F (approximate % relative to control)	5F (approximate % relative to control)
Hb (g/dL)	4	15.8	15.1	14.8*	14.2*** (10% less)	14.7** (7% less)
	13	15.1	14.9	14.3	12.3** (19% less)	13.4 (11% less)
PCV (%)	4	47.1	44.9	44.8	43.0*** (9% less)	43.9** (7% less)
	13	42.0	41.6	40.6	35.1** (16% less)	37.8 (10% less)
MCV (fL)	4	57.7	56.6	55.1	54.2** (6% less)	55.2 (4% less)
	13	53.9	53.8	51.2	48.4** (10% less)	49.4* (8% less)
MCH (pg)	4	19.4	19.2	18.2*	17.9*** (8% less)	18.5 (5% less)
	13	19.4	19.2	18.0*	16.8*** (13% less)	17.8** (10% less)
Platelets (1000/cmm)	4	1008	1102	1109	1192 (18% more)	1037
	13	1021	997	1045	1265*** (24% more)	1116 (9% more)

* P<0.05
** P<0.01
*** P<0.001

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

8. Clinical Chemistry: Slight changes of total bilirubin, unsaturated iron binding capacity (UIBC), total iron binding capacity (TIBC), and serum iron level were noted and these results were summarized in Table 5 on page 55 in volume 1.17. This table is attached below (Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively).

Clinical chemistry Table 5, Appendix 5

Measurement code (unit)	Week	Males				
		1M	2M	3M	4M (approximate % relative to control)	5M (approximate % relative to control)
T BILI (umol/L)	4	1.7	2.1	1.7	2.9*** (71% more)	2.2 (29% more)
	13	1.6	2.0	1.8	2.5** (56% more)	1.9 (19% more)
Glucose	4	-	-	-	-	↓
Fe (umol/L)	5	38.2	45.3	66.5*	53.1	36.8
	14	30.6	50.1*	60.7***	98.7*** (223% more)	62.0*** (103% more)
UIBC (umol/L)	5	65.0	56.5	44.2	78.6 (21% more)	82.4* (27% more)
	14	55.2	37.2	39.2	13.6*** (75% less)	36.6 (34% less)
TIBC (umol/L)	5	105.1	104.8	110.2	131.7 (25% more)	119.2 (13% more)
	14	85.8	86.9	96.5	112.3*** (31% more)	98.6 (15% more)

- = No change
↑ = slight increase
↓ = slight decrease
* P<0.05
** P<0.01
*** P<0.001

Measurement code (unit)	Week	Females				
		1F	2F	3F	4F (approximate % relative to control)	5F (approximate % relative to control)
T BILI (umol/L)	13	2.2	2.4	2.6	3.2* (45% more)	2.9 (32% more)
K	4	-	↑	↑	↑	↑
Fe (umol/L)	5	72.4	83.1	62.7	38.7** (47% less)	46.6* (36% less)
	14	59.2	69.1	82.3	65.2	75.5
UIBC (umol/L)	5	31.3	25.6	47.5	92.5*** (196% more)	72.9** (133% more)
	14	40.3	32.6	18.2**	66.9* (66% more)	33.2
TIBC (umol/L)	5	103.7	106.5	110.9	131.2*** (27% more)	119.5* (15% more)
	14	99.5	101.7	99.6	132.1*** (33% more)	108.7 (9% more)

- = No change
↑ = slight increase
↓ = slight decrease
* P<0.05
** P<0.01
*** P<0.001

Serum gastrin level was increased in treatment groups and these results were summarized in a table on page 57 in volume 1.17. This table is attached below.

Mean serum gastrin concentrations (24 h after dose) ±SD after 12 weeks' treatment

	Males					Females				
	1	2	3	4	5	1	2	3	4	5
Gastrin (pg/ml)	100 ±20	140 ±50	470 ±330	1300 ±600	890 ±460	78 ±15	150 ±60	810 ±440	2000 ±600	1100 ±600

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

There were no treatment related changes in the plamsa thyroid hormone level (T_3 and T_4).

9. Urinalysis: There were no treatment related changes.

10. Organ Weights: Increases in the stomach, liver, and kidney weights were found mainly in the high dose group and omeprazole group. These results were summarized in Table 7 on pages 57 and 58 in volume 1.17. This table is attached below.

Organ weights Table 7, Appendix 9

Organ weight(g) #	Males				
	1M	2M	3M (approximate % relative to control)	4M (approximate % relative to control)	5M (approximate % relative to control)
Stomach	1.978	2.462**	2.964*** (50% more)	2.902*** (47% more)	2.746*** (39% more)
Liver	12.941	13.092	14.418	14.732* (14% more)	14.613 (13% more)
Kidney	2.374	2.586	2.515	2.623 (10% more)	2.639 (11% more)
Heart	-	-	†	†	†

APPEARS THIS WAY
ON ORIGINAL

Organ weight(g) #	Females				
	1F	2F	3F(approximate % relative to control)	4F (approximate % relative to control)	5F (approximate % relative to control)
Stomach	1.470	1.724*	1.879***(28% more)	2.059*** (40% more)	1.998***(36% more)
Liver	8.206	8.148	8.514	10.525*** (28% more)	9.090 (11% more)
Kidney	1.583	1.620	1.560	1.709 (8% more)	1.812* (14% more)
Heart	-	-	-	↑	↑

organ weights are adjusted to overall mean necropsy brain weight
-= No change
↑= slight increase
↓= slight decrease
* P<0.05
** P<0.01
*** P<0.001

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

11. Gross Pathology: Dark focus in the stomach was noted in 6 high dose males.

12. Histopathology: Histopathological examination revealed treatment related changes in the stomach and kidney and these results were summarized in a table on page 59 in volume 1.17. This table is attached below.

Incidence of treatment-related findings in the stomach

Sex		Males					Females				
Dose group		1	2	3	4	5	1	2	3	4	5
No. of animals		10	10	10	10	10	10	10	10	10	10
STOMACH											
Chief cell eosinophilia	**Total**	-	1	7	10	9	-	-	5	10	9
	Minimal	-	1	6	1	8	-	-	4	3	7
	Slight	-	-	-	6	1	-	-	1	6	2
	Moderate	-	-	1	3	-	-	-	-	1	-
Acanthosis	**Total**	-	-	-	7	2	-	-	-	2	-
	Minimal	-	-	-	5	2	-	-	-	2	-
	Slight	-	-	-	2	-	-	-	-	-	-
Hyperkeratosis	**Total**	-	-	-	8	2	-	-	-	2	-
	Minimal	-	-	-	2	2	-	-	-	2	-
	Slight	-	-	-	6	-	-	-	-	-	-

Incidence of renal basophilic tubules

Sex		Males					Females				
Dose group		1	2	3	4	5	1	2	3	4	5
No. of animals		10	10	10	10	10	10	10	10	10	10
KIDNEYS											
Basophilic cortical tubules	**Total**	3	2	5	8	7	2	1	2	8	5
	Minimal	3	1	4	4	4	2	1	2	2	4
	Slight	-	-	1	3	3	-	-	-	3	1
	Moderate	-	1	-	1	-	-	-	-	3	-

Groups 1, 2, 3, 4, and 5 represent control, 14, 69, 280 mg/kg/day of H199/18, and 140 mg/kg/day of omeprazole, respectively.

13. Toxicokinetics: In general, plasma levels of H 199/18 were higher in female than in males. This difference was more obvious after 3 month treatment. Plasma levels of H 199/18 were accumulated over time. These differences were also observed in the omeprazole group. These results were summarized in a table on page 52 in volume 1.17. This table is attached below (1 µmol ≈ 0.35 mg).

- **Dose levels and exposure to H 199/18 or omeprazole**

Compound	Dose		Median (range) C_{max} (μmol/l)		Median (range) AUC(0-2 h) (μmol·min/l)	
	μmol/kg/day	mg/kg/day	Day 1	3 months	Day 1	3 months
Males						
H 199/18	40	14	<0.10	0.25	NC	NC
H 199/18	200	69	0.64	1.62	NC	67
H 199/18	800	280	3.93	18.8	176	909
Omeprazole	400	140	4.93	6.09	227	533
Females						
H 199/18	40	14	0.13	0.53	NC	24
H 199/18	200	69	0.66	12.8	32	389
H 199/18	800	280	10.4	57.6	477	3880
Omeprazole	400	140	7.16	34.5	372	2110

NC = Not calculated

In summary, H 199/18 was tested orally in Sprague-Dawley rats at 0, 14, 69, and 280 mg/kg/day for three months. Decreases (4-19%) in the mean concentrations of hemoglobin (Hgb), packed cell volume (PCV), mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH), and increase (9-34%) in the mean platelet count were found in the high dose group and omeprazole group as compared to the control. Histopathological changes were mainly in the high dose group and these changes included chief cell eosinophillia, acanthosis, and hyperkeratosis in stomach and basophilic coritcal tubules in kidney.

DOG:

3-Month Oral (gavage) Toxicity Study in Dogs (97103)

Testing Laboratories: Sponsor's laboratories:

Safety Assessment, Leics, England
Safety Assessment, Sodertalje, Sweden

Study Start and Completion Date: June 9, 1997 and September 21, 1998

GLP Requirement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Male: 9.0-14.6 kg, 7-10 months old
Female: 7.3-11.8 kg, 7-10 months old
Beagle dogs

Drug Batch No.: 602/97

Methods: To evaluate the toxicity of H 199/18 magnesium in dogs, dogs (3/sex/group) were given H 199/18 at 0, 0.65, 5.5 and 28 mg/kg/day for 3 months by oral gavage. Omeprazole magnesium at 28 mg/kg/day was also given as positive control. Both H 199/18 and omeprazole were given in suspension in 0.5% hydroxy- propylmethyl cellulose. The basis of dose selection was not provided in this submission. The clinical sign of toxicity was observed daily. The body weights were recorded weekly. Food consumption was recorded daily. Ophthalmic examinations were conducted before treatment started and during week 12. ECGs were performed pretest, on day 2 and during weeks 5 and 12. All recordings were taken ~20 minutes after dosing. Hematology, clinical chemistry and urinalysis were determined before treatment and during weeks 4 and 12. The blood samples were taken before each dosing during weeks 4 and 12. Gastrin level was determined before treatment and during week 13. All animals were necropsied at termination and organs were weighed. Histopathological examination was conducted in all animals necropsied at termination. Plasma level of test drug was determined on days 1, 29 and 84.

Results:

1. Clinical Signs: Head nodding, unsteady gait and subdued behavior were observed in the high dose males. Red/brown urine was noted in the high dose animals and the animals treated with omeprazole.

2. Mortality: There were no deaths.

3. Body Weight: The mean initial and final body weights in the control animals were 12.7 kg and 13.5 kg (males) or 9.1 and 11.5 kg (females), respectively. There were no treatment related changes.

4. Food Consumption: There were no treatment related changes.

5. Ophthalmoscopy: There were no treatment related alterations observed during the study.

6. ECG: There were no treatment related changes.

7. Hematology: There were no clear treatment related changes.

8. Clinical Chemistry: Cholesterol level was increased by 25-40% mainly in the mid and high dose groups and omeprazole group. Serum gastrin level was increased in all treatment groups as compared to the control and this information was summarized in a table on page 36 in volume 1.18. This table is attached below.

Gastrin Levels at Week 13

Group	Compound	Dose Level ($\mu mol \cdot kg^{-1} \cdot day^{-1}$) [$mg \cdot kg^{-1} \cdot day^{-1}$]	Basal ($pg \cdot ml^{-1}$)		Food Stimulated ($pg \cdot ml^{-1}$)			
			-30 min	0 min	30 min	60 min	90 min	180 min
1	Vehicle	0	a	a	53 ±16	a	a	a
2	H 199/18	1.9 [0.65]	a	a	170 ±140	130 ±90	110 ±90	120 ±60
3	H 199/18	16 [5.5]	320 ±300	310 ±270	700 ±210	650 ±390	610 ±360	500 ±270
4	H 199/18	80 [28]	240 ±210	140 ±110	620 ±290	500 ±230	460 ±230	520 ±380
5	Omeprazole	80 [28]	180 ±150	160 ±140	470 ±210	410 ±160	370 ±110	340 ±140

Individual gastrin levels <50 $pg\ ml^{-1}$ are set at 25 $pg\ ml^{-1}$ in calculation of the means.

a = No mean levels were estimated when more than 2 out of 6 concentrations were <50 $pg\ ml^{-1}$.

9. Urinalysis: There were no treatment related changes.

10. Organ Weights: The relative stomach and liver weights were increased in the treatment groups and this information was summarized in a table on page 36 in volume 1.18. This table is attached below.

APPEARS THIS WAY
ON ORIGINAL

Relative Mean Organ Weights (% of body weight)										
	1M	2M	3M	4M	5M	1F	2F	3F	4F	5F
Stomach	0.99	1.15	1.48	1.36	1.61	1.02	1.14	1.34	1.23	1.46
(% difference)	-	+16	+49	+37	+63	-	+12	+31	+21	+43
Liver	3.42	3.52	3.66	3.80	3.88	3.49	3.58	3.62	3.77	4.12
(% difference)	-	+3	+7	+11	+13	-	+3	+4	+8	+18

11. Gross Pathology: There were no treatment related changes.

12. Microscopic Pathology: Histopathological changes were limited in the stomach and the incidence and severity of these changes were summarized in a table on page 37 in volume 1.18. This table is attached below.

Group	1M	2M	3M	4M	5M	1F	2F	3F	4F	5F
No. Animals	3	3	3	3	3	3	3	3	3	3
Mucosal hyperplasia	0	1	3	2	2	0	0	3	3	3
Mean severity	-	1.0	1.7	1.5	2.0	-	-	1.0	1.3	1.7
Chief cell atrophy	0	1	3	3	3	0	1	3	2	3
Mean severity	-	1.0	1.3	1.3	2.0	-	1.0	1.7	1.0	2.0
Mucosal fibrosis	0	1	3	3	3	1	0	2	2	3
Mean severity	-	1.0	1.0	1.0	2.0	1.0	-	1.0	1.5	1.0
Inflammatory cells Lamina propria	0	1	0	2	3	0	0	0	0	1
Mean severity	-	1.0	-	1.0	2.3	-	-	-	-	1.0
Focal necrosis, gastric glands	0	0	0	1	2	0	0	1	0	1
Mean severity	-	-	-	1.0	2.0	-	-	1.0	-	2.0
Sum of mean severity scores	**0**	**4.0**	**4.0**	**5.8**	**10**	**1.0**	**1.0**	**4.7**	**4.8**	**7.7**
Total severity scores	**0**	**4**	**12**	**13**	**27**	**1**	**1**	**11**	**9**	**17**

13. Plasma Level of Test Drug: There was no significant difference in the plasma levels of the test drugs between days 1, 29, and 84. The overall mean values on days 1, 29, and 84 were provided. The maximum plasma levels (C_{max}) of the test drug were 2.93 ± 1.7, 17.9 ± 6.3 and 75.4 ± 24.1 µmol/l in the low, mid and high dose groups, respectively. The AUC values were 169 ± 85, 1100 ± 410 and 5140 ± 1890 µmol.min/l in the low, mid and high dose groups, respectively. The C_{max} and AUC of omeprazole were 65.5 ± 16.8 µmol/l and 5310 ± 2030 µmol.min/l, respectively (1 µmol ≈ 0.35 mg).

In summary, H 119/18 was tested in dogs at 0, 0.65, 5.5, and 28 mg/kg/day for 3 months by oral gavage. The major treatment related changes were histopathological changes in the stomach including mucosal fibrosis and hyperplasia, Chief cell atrophy and focal necrosis in all treatment groups including omeprazole. Serum gastrin level was increased in all treatment groups. The stomach was the target organ of toxicity.

REPRODUCTIVE TOXICITY:

A Segment II Oral Teratological Study of H 199/18 in Rats (97469)

Testing Laboratory: Astra AB, Safety Assessment
Sodertalje, Sweden

Study Start and Completion Dates: February 23, 1998 and
April 23, 1999

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Females (190-310 g, ~10 weeks old)
Sprague-Dawley rats

Methods: To evaluate the teratological potential of H 199/18, H 199/18 was given to pregnant female rats by oral gavage at 0, 14, 69, and 280 mg/kg/day during Gestation Days 6 through 16. An additional group of pregnant rats was treated with omeprazole at 140 mg/kg/day. The dose selection was based on the results of a dose ranging study in pregnant rats (study #97207). In this study, the high dose of 280 mg/kg/day markedly decreased body weight gain during first three days of treatment. Therefore, the high dose of 280 mg/kg/day was selected for the current study. All dams were observed daily for clinical signs of toxicity and mortality. Body weights and food consumption were recorded daily. All pregnant rats were sacrificed on Gestation Day 21 for assessment of fetal viability, weight and morphology.

Results: There were no deaths in this study. "Ploughing" with nose in the cage bedding was noted after dosing in one mid dose rat and most of the high dose rats. The body weight gain during the treatment period (pregnant days 6-16) was decreased by ~8% and 20% in the mid and high dose groups, respectively, as

compared to the control. This was consistent with the decreased food consumption in the high dose group (18.9-25 g/day) as compared to the control (21.3-27.6 g/day).

Number of corpora lutea and implantations were not adversely affected. Fetal examination did not reveal any significant treatment related changes in number of dead and live fetuses, body weight of the live fetuses, placental weight and sex ratio. The results were summarized in Table 5 on pages 123 and 124. This table is attached below.

Table 5 Cesarean data

LAC996-07/06 LAC996P/1 ** ** RUN DATE: 11/01/99 TIME: 11:08

INTERGROUP COMPARISON OF PREGNANCY DATA PAGE: 1

T-TEST:- GROUP 1 COMPARED WITH ALL OTHER GROUPS (* P < 0.05 ** P < 0.01, TWO SIDED)

	GROUP 1 0 UMOL/KG	GROUP 2 B 199/18 40 UMOL/KG	GROUP 3 B 199/18 200 UMOL/KG	GROUP 4 B 199/18 800 UMOL/KG	GROUP 5 OMEPRAZOLE 400 UMOL/KG
NUMBER OF PREGNANCIES	23	22	23	24	24
NUMBER OF FEMALES WITH LITTERS (TOTAL)	23	22	23	24	24
(%)	100.0	100.0	100.0	100.0	100.0
NUMBER OF INTERCURRENT DEATHS	0	0	0	0	0
NUMBER OF FEMALES AT TERM WITH RESORPTIONS ONLY	0	0	0	0	0
NUMBER OF FEMALES WITH ABORTIONS	0	0	0	0	0
GRAVID UTERUS WEIGHT (G) (MEAN)	82.2 WW	88.5	80.5 WW	79.5 WW	80.2
(S.D.)	10.2	10.8	8.7	11.1	10.3
(NUMBER OF VALUES)	(22)	(22)	(22)	(23)	(24)
TOTAL NUMBER OF LIVE FOETUSES	276	290	274	281	280
% MALES	51.8	46.6	48.2	53.4	52.9
LITTER WEIGHT (G) (MEAN)	62.4	67.3	61.3	61.8	61.0
(S.D.)	16.6	8.0	7.1	8.6	8.5
(NUMBER OF VALUES)	(23)	(22)	(23)	(24)	(24)
FOETAL WEIGHT (G) (MEAN)	5.2	5.1**	5.1	5.2	5.2
(S.D.)	0.4	0.4	0.5	0.4	0.4
(NUMBER OF VALUES)	(275)	(290)	(274)	(281)	(280)

WW The weight of one gravid uterus is missing

Appears this way on original

29(193)

INTERGROUP COMPARISON OF CAESARIAN DATA AT TERMINATION PAGE: 2

T-TEST:- GROUP 1 COMPARED WITH ALL OTHER GROUPS (* P < 0.05 ** P < 0.01, TWO SI

	GROUP 1 0 UMOL/KG	GROUP 2 H 199/18 40 UMOL/KG	GROUP 3 H 199/18 200 UMOL/KG	GROUP 4 H 199/18 800 UMOL/KG	GROUP 5 OMEPRAZOLE 400 UMOL/KG
NUMBER OF CORPORA LUTEA					
(MEAN)	14.2	14.2	13.8	13.1*	14.8
(S.D.)	1.9	1.6	1.9	1.5	2.2
(NUMBER OF VALUES)	(23)	(22)	(23)	(24)	(24)
NUMBER OF IMPLANTATIONS (TOTAL)	286	297	286	290	298
(MEAN)	12.4	13.5	12.4	12.1	12.4
(S.D.)	3.0	1.8	1.4	1.7	2.0
(NUMBER OF VALUES)	(23)	(23)	(23)	(24)	(24)
PRE-IMPLANTATION LOSS (ADJUSTED[a]) (%)	12.3	6.8	9.8	7.9	11.3
PROPORTION OF LITTERS AFFECTED	13/23	9/22	17/23	13/24	13/24
NUMBER OF FOETUSES (TOTAL)	276	290	274	281	280
(MEAN)	12.0	13.2	11.9	11.7	11.7
(S.D.)	2.9	1.6	1.6	1.7	1.9
% OF IMPLANTATIONS	96.5	97.6	95.8	96.9	94.0
INTRA-UTERINE DEATHS					
EARLY (TOTAL)	9	7	10	9	17
(MEAN)	0.4	0.3	0.4	0.4	0.7
(S.D.)	0.7	0.6	0.6	0.6	0.7
LATE (TOTAL)	1	0	2	0	1
(MEAN)	0.0	0.0	0.1	0.0	0.0
(S.D.)	0.2	0.0	0.3	0.0	0.2
TOTAL (TOTAL)	10	7	12	9	18
(MEAN)	0.4	0.3	0.5	0.4	0.8
(S.D.)	0.7	0.6	0.6	0.6	0.7
POST IMPLANTATION LOSS (%)	3.5	2.4	4.2	3.1	6.0
PROPORTION OF LITTERS AFFECTED	8/23	6/22	11/23	8/24	16/24

a) If the number of corpora lutea was less than the number of implantations, the number of corpora lutea was adjusted to equal the number of implantations

There were no treatment related changes in the major malformations. One control fetus had right sided aortic arch, bilateral absence of renal papilla and increased dilation of the urethra (bilateral). Another control fetus had agnathia. These two fetuses were from two separate litters. Two fetuses from same litter in omeprazole group showed bilateral undescended testes. There were no clear treatment related changes in the minor defects and variants. These results are summarized in the following table.

Major Malformations

Parameter	Control	14 mg/kg/day	69 mg/kg/day	280 mg/kg/day	Omeprazole
# Fetuses (litters) External and Visceral	276 (23)	290 (22)	274 (23)	281 (24)	280 (24)
Agnathia	1 (1)				
Right sided aortic arch	1 (1)				
Undescended testes					2 (1)
# Fetuses (litters) Skeletal	137 (23)	144 (22)	137 (23)	139 (24)	140 (24)
No major defects were detected.					

In summary, in the Segment II teratological reproductive toxicity study in rats, H 199/18 was given to rats by oral gavage at 0, 14, 69, and 280 mg/kg/day during days 6 through 16. Maternal toxicity was evidenced by decreased body weight gain in the high (20%) dose group. There were no significant treatment related changes on number of corpora lutea, implantations, number of dead and live fetuses, body weight of the live fetuses, placental weight, and sex ratio. There were no treatment related changes in the major malformations. H 199/18 was not teratogenic in this study.

A Segment II Oral Teratological Study of H 199/18 in Rabbits (98498)

Testing Laboratory: AstraZeneca, R&D
Sodertal JE, Sweden

Study Start and Completion Dates: January 5, 1999 and July 30, 1999

GLP and QAU Compliance Statement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Females (2.7-4.1 kg, ~4 months old)
New Zealand White rabbits

Drug Batch No: 1300/98

Methods: To evaluate the teratological potential of H 199/18, H 199/18 was given to pregnant female rabbits by oral gavage at 0, 20, 80, and 250 µmol/kg/day (0, 6.9, 28, and 86 mg/kg/day) during

Gestation Days 6 through 16. An additional group of pregnant rabbits was treated with omeprazole at 80 µmol/kg/day (28 mg/kg/day). The dose selection was based on dose ranging studies in rabbits (studies #97325, 98107, and 98344). In study #97325, dose of 200 µmol/kg/day (~69 mg/kg/day) was tolerated. In study # 98344, oral dose (gavage) of H 199/18 was given to pregnant rabbits during pregnant days 6 to 18 at 0, .250, and 300 µmol/kg/day (86 and 103 mg/kg/day) and decreased body weight was noted during first two days of treatment (days 6-8) in both groups (-46 g and 78 g in 250 and 300 µmol/kg/day groups, respectively, and control group had 56 g gain during the same period). In study #98107, oral dose (gavage) of H 199/18 was given to pregnant rabbits during pregnant days 6 to 15 at 0, 400, and 800 µmol/kg/day (138 and 276 mg/kg/day). H 199/18 was lethal at doses of 400 µmol/kg/day or higher. Based on these results, the selection of doses of 20, 80, and 250 µmol/kg/day appeared adequate. In the current study, all dams were observed daily for clinical signs of toxicity and mortality. Body weights and food consumption were recorded daily. All pregnant rabbits were sacrificed on gestation Day 29 for assessment of fetal viability, weight and morphology.

Results: There were no deaths in this study. Soft feces was observed more frequently in the high dose group. The body weight was decreased by ~70 g in the high dose group and ~17 g in the omeprazole group. The body weight gain during the treatment period was decreased by 30.8%, 79.8% in the in the mid and high dose groups, and by 48% in omeprazole group, respectively, as compared to the control. This was consistent with the decreased food consumption in the mid (96.8-135.7 g/day) and high (85.9-110.6 g/day) groups and in the omeprazole group (99.9-129.1 g/day) as compared to the control (129-154.4 g/day).

Number of corpora lutea and implantations were not adversely affected. Fetal examination did not reveal any significant treatment related changes in number of dead and live fetuses, placental weight and sex ratio. A slight decrease in the fetal body weight was noted in the high dose group. The results were summarized in Table 4 on pages 284, 285, and 286 in volume 1.20. This table is attached below.

Table 4 Intergroup comparison of caesarian data

Disposition of animals						REP1
Group	:	1	2	3	4	5
Test article	:	Control	Low Dose	Medium Dose	High Dose	Comp Dose
Dose level (µmol/kg)	:	0	20 µmol/kg	80 µmol/kg	250 µmol/kg	80 µmol/kg

Number of females:	Group 1	Group 2	Group 3	Group 4	Group 5
In group	20	20	20	20	20
Not pregnant (%)	0 (0.0)	2 (10.0)	0 (0.0)	0 (0.0)	1 (5.0)
Died/killed	0	0	0	0	0
Survived to scheduled kill	0	2	0	0	1
Pregnant (%)	20 (100.0)	18 (90.0)	20 (100.0)	20 (100.0)	19 (95.0)
Died/killed/aborted	0	0	0	0	0
with total resorptions	0	0	0	0	0
with live foetuses at scheduled kill	20	18	20	20	19

APPEARS THIS WAY
ON ORIGINAL

Table 4 Intergroup comparison of caesarian data, continued

Group mean uterine / implantation data

Group	:	1	2	3	4	5
Test article	:	Control	Low Dose	Medium Dose	High Dose	Comp Dose
Dose level (µmol/kg)	:	0	20 µmol/kg	80 µmol/kg	250 µmol/kg	80 µmol/kg

	Group 1	Group 2	Group 3	Group 4	Group 5
Number of females with implantations at scheduled kill	20	18	20	20	19
Number of corpora lutea	217	198	215	219	205
Mean number per female	10.9	11.0	10.8	11.0	10.8
Standard deviation	(1.6)	(2.0)	(2.1)	(2.5)	(2.0)
Number of implantations	195	177	209	194	181
Mean number per female	9.8	9.8	10.5	9.7	9.5
Standard deviation	(2.3)	(2.3)	(2.1)	(2.9)	(3.0)
Mean % pre-implantation loss	10.7	9.9	2.8	10.1	13.7
Number of early embryo/foetal deaths	16	8	8	17	9
Number of late embryo/foetal deaths	8	2	13	5	3
Number of dead foetuses	0	0	1	0	0
Mean % post-implantation loss	14.8	6.2	10.2	11.7	6.3
Number of live foetuses	171	167	187	172	169
Mean number per female	8.6	9.3	9.4	8.6	8.9
Standard deviation	(2.9)	(2.4)	(2.0)	(2.9)	(2.9)
Mean % of implantations	85.2	93.8	89.8	88.3	93.7

Table 4 Intergroup comparison of caesarian data, continued

Group mean litter weights (g) / foetal data

Group	:	1	2	3	4	5
Test article	:	Control	Low Dose	Medium Dose	High Dose	Comp Dose
Dose level (μmol/kg)	:	0	20 μmol/kg	80 μmol/kg	250 μmol/kg	80 μmol/kg

	Group 1	Group 2	Group 3	Group 4	Group 5
Number of females with live foetuses at scheduled kill	20	18	20	20	19
Number of live foetuses	171	167	187	172	169
Mean number per female	8.6	9.3	9.4	8.6	8.9
Standard deviation	(2.9)	(2.4)	(2.0)	(2.9)	(2.9)
Number of male foetuses	88	80	100	82	86
Number of female foetuses	83	87	87	90	83
Mean % male foetuses	52.9	48.8	52.8	48.8	48.2
Mean litter weight	316.9	339.1	338.0	291.2	323.8
Standard deviation	(92.2)	(71.1)	(71.1)	(67.3)	(9.6)
Mean foetal weight	38.0	37.4	36.3	35.5	37.0
Standard deviation	(4.3)	(4.8)	(3.6)	(5.8)	(3.6)
Mean foetal weight - males only	38.5	38.0	37.0	35.5	37.3
Standard deviation	(4.2)	(4.4)	(4.3)	(6.0)	(2.8)
Mean foetal weight - females only	36.8	36.9	35.9	34.0	36.3
Standard deviation	(5.1)	(5.3)	(4.0)	(4.8)	(4.4)

There were no clear treatment related changes in the major malformations. These results are summarized in the following table.

Major External and Visceral Malformations

Parameter	Control	6.9 mg/kg/day	28 mg/kg/day	86 mg/kg/day	Omeprazole
# Fetuses (litters)	171 (20)	167 (18)	188 (20)	172 (20)	169 (19)
Visceral					
Microphthalmia	1 (1)				
Cleft palate	1 (1)			1 (1)	
Aortic and pulmonary arches: persistent truncus arteriosus		1 (1)			
Enlarged heart	1 (1)			1 (1)	
Interventricular septum (incomplete)	1 (1)	1 (1)			
Increased pelvic cavitation in kidney		1 (1)			
Dilated ureter	1 (1)	1 (1)			
External					
Multiple cranio-facial abnormality	1 (1)				
Thoracoschisis		1 (1)			
Spina bifida		1 (1)			

Major Skeletal Malformations

Parameter	Control	6.9 mg/kg/day	28 mg/kg/day	86 mg/kg/day	Omeprazole
# Fetuses (litters)	171 (20)	167 (18)	188 (20)	172 (20)	169 (19)
Skull:					
Acrania	1 (1)				
Fused nasal		1 (1)			
Fused frontal		1 (1)		1 (1)	
Fused parietal		1 (1)		1 (1)	
Short mandible	1 (1)				
Vertebra					
Fused centra		1 (1)			
Fused neural arch				1 (1)	
Thoracic vertebra					
Absent neural arch			1 (1)	1 (1)	
Lumbar vertebra					
Fused centra		1 (1)			
Absent neural arch		1 (1)			
Rib					
Fused rib			1 (1)	1 (1)	
Absent rib		1 (1)	1 (1)		
Sternum					
Fused sternum	1 (1)		1 (1)		2 (2)
Forelimb					
Absent pollex	1 (1)				

There were no clear treatment related changes in the variants and minor malformations.

In summary, in the Segment II teratological reproductive toxicity study in rabbits, H 199/18 was given to rabbits by oral gavage at 0, 6.9, 28, and 86 mg/kg/day during days 6 through 16. Maternal toxicity was evidenced by decreased body weight (high dose) and body weight gain in the mid and high dose groups. There were no significant treatment related changes on number of corpora lutea, implantations, number of dead and live fetuses, body weight of the live fetuses, placental weight, and sex ratio. There were no treatment related changes in the major malformations. H 199/18 was not teratogenic in this study.

MUTAGENICITY:

Salmonella Typhimurium Gene Reverse Mutation Test of H199/18 Sodium: Ames Test
(Report # T2817)

Testing Laboratories: Astra Hassle AB,
Sodertalje, Sweden.

Study Started: January 10, 1994

Study Completed: May 19, 1994

GLP Requirements: A statement of compliance with the GLP regulations and quality assurance unit was included.

Cells Employed: Salmonella Typhimurium strains TA1535, TA100, TA1538, TA98, TA1537.

Concentrations Employed: 119, 398, 1190, 3980, and 11900 µg/plate (or 0.325, 1.08, 3.25, 10.8, and 32.5 µmol/plate).

Basis of Dose Selection: The dose selection was based on the first dose-range toxicity/mutagenicity study with all strains, where doses of 119-11900 µg/plate of H199/18 sodium were tested. Precipitate was formed at concentrations of 3980, and 11900 µg/plate. In the second, and third mutagenicity studies

concentrations of 60-6400 µg/ plate (or 0.164-17.5 µmol) were tested.

Solvent Control: Sterile double-distilled water.

Positive Controls: Sodium azide (0.5 µg/plate), 2-nitrofluorene (0.5 µg/plate), 9-aminoacridine (75 µg/plate) 2-aminoanthracene (2 µg/plate).

Source of Metabolic Activation: Rat liver microsome S-9 fraction.

Drug Batch No: 600/93.

Criteria of Genotoxic Effect: If there was a significant increase (using a Dunnett's test for multiple comparisons with $p \leq 0.01$) in the number of revertant colonies in the presence of the test compound under any of the experimental conditions and the number of revertant colonies in the solvent control were in the normal range, while the positive controls invoked marked increase, the test was considered positive.

Methods: The 5 salmonella tester strains in plates were exposed to vehicle, H199/18 sodium, or positive controls. The cells were incubated for approximately 48-72 hours at 37° C on selective minimal-glucose agar in both the presence and absence of S-9 fraction. Colonies were counted at the end of study either manually or using the automated colony counter.

Results: Total of 3 studies were conducted. In the first dose-range study, 119-11900 µg/plate (or 0.325-32.5 µmol) were used to check the toxicity as well as mutagenicity. In the second and third studies, concentrations of 60-6000 µg/plate (0.164-16.4 µmol) and 64-6400 µg/plate (0.175-17.5 µmol) were used. In all 3 studies, with or without metabolic activation, H199/18 sodium was not mutagenic in any of the tester strains at doses ranging from 119-11900 µg/plate. The drug did show toxicity at ≥5.83 µmol/ plate (2140 µg/plate) in the presence and absence of metabolic activation system. A significant increase in mutant colonies was observed in all the microbial strains employed with positive controls (with or without S-9 mix). These studies indicate that H199/18 sodium does not have a mutagenic potential in the Ames test.

Experiment No. 3	Final concentration in culture medium[1]		% Mitotic Index[2] (compared with control)	% Cells with[2,3] aberrations (excluding gaps)
	(mmol/L)	(mg/mL)		
Negative control (DMSO)	0.5% v/v		100	0.3
H199/18	1.30	0.4492	94	4.0***
	1.35	0.4664	61	10.5***
	1.40	0.4837	57	9.5***
	1.45	0.5010	36	12.0***
H199/19	1.40	0.4837	40	13.5***
	1.45	0.5010	58	9.0***
	1.55	0.5355	46	10.0***
Omeprazole	1.35	0.4664	56	8.0***
	1.40	0.4837	43	12.0***
	1.45	0.5010	46	8.0***
H 225/20	0.50	0.1847	79	2.0*
	0.55	0.3032	75	7.0***
	0.60	0.2216	58	8.5***
Positive control NQO	2.5 µg/mL		-	11.0***

[1] H 199/18, H 199/19 and omeprazole were formulated as the sodium salts, but all concentrations are expressed as the neutral form (H 199/18, H 199/19 and omeprazole).
[2] Mean of duplicate or six (negative control) cultures.
[3] Fisher's exact test used. * P≤0.05 ** P≤0.01 *** P≤0.001

In conclusion, H 199/18 was inducer of chromosomal aberrations in this test system and omeprazole and its enantiomers induced aberrations to a similar degree.

In Vivo Chromosome Aberration Test in Rat Bone Marrow Cell With H 199/18 (SR98457-01)

Testing Laboratory: [redacted]

Dates Started and Completed: December 7, 1998 and May 24, 1999

Compliance with GLP and OAU Requirement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Male (230-265 g, ~8 weeks old)
Female (188-220 g, ~8 weeks old)
Crl:CD BR (CD) rats

Methods: To examine the potential mutagenic effect of H 199/18, an *in vivo* chromosome aberration test was conducted. A single dose of H 199/18 was given to rats (6/sex/group, 12/sex in control and high dose groups) by oral gavage at 0, 487, 975, and 1950 mg/kg in males and 0, 438, 875, and 1750 mg/kg in females. The dose selection was based on the results of a dose ranging study. In this study, the high dose of 2000 mg/kg/day produced clinical signs of toxicity including convulsion, eye closure, abnormal breathing, and gasping. Positive (cyclophosphamide, 40 mg/kg) control was also tested. The treated animals were sacrificed at 24 and 48 hours after dosing and bone marrow was collected. The chromosomal aberrations were then observed and compared between the control and test groups. To arrest cells at metaphase, colchicine (2 mg/kg) was intraperitoneally injected at ~2 hours before sacrifice.

Results: H 199/18 did not significantly increase the frequency of the chromosomal aberration as compared to the control. However, the positive controls significantly increased it.

In conclusion, the results suggest that H 199/18 was not mutagenic in this test system.

Addendum: A toxicokinetic study was conducted in rats to determine the plasma level of H 199/18 (study #99550). In this study, rats were treated with H 199/18 by a single oral dose (gavage) at 0, 1000, and 2000 mg/kg (males) or 0, 900, and 1800 mg/kg (females). The maximum plasma levels of H 199/18 were ~64 and 119 µmol/l for 1000 and 2000 mg/kg males or 281 and 437 µmol/l for 900 and 1800 mg/kg females, respectively. AUC values were 6670 and 16100 µmol.min/l for 1000 and 2000 mg/kg males or 49900 and 107000 µmol.min/l for 900 and 1800 mg/kg females, respectively. The percentage of protein binding was 68-83%.

Micronucleus Test of H 199/18 in Mice by Oral Gavage

(97484)

Testing Laboratory: Sponsor's laboratory

Dates Started and Completed: March 19, 1998 and May 28, 1998

Methods: To examine the potential mutagenic effect of H 199/18, an *in vivo* chromosome aberration test was conducted. A single dose of H 199/18 was given to rats (6/sex/group, 12/sex in control and high dose groups) by oral gavage at 0, 487, 975, and 1950 mg/kg in males and 0, 438, 875, and 1750 mg/kg in females. The dose selection was based on the results of a dose ranging study. In this study, the high dose of 2000 mg/kg/day produced clinical signs of toxicity including convulsion, eye closure, abnormal breathing, and gasping. Positive (cyclophosphamide, 40 mg/kg) control was also tested. The treated animals were sacrificed at 24 and 48 hours after dosing and bone marrow was collected. The chromosomal aberrations were then observed and compared between the control and test groups. To arrest cells at metaphase, colchicine (2 mg/kg) was intraperitoneally injected at ~2 hours before sacrifice.

Results: H 199/18 did not significantly increase the frequency of the chromosomal aberration as compared to the control. However, the positive controls significantly increased it.

In conclusion, the results suggest that H 199/18 was not mutagenic in this test system.

Addendum: A toxicokinetic study was conducted in rats to determine the plasma level of H 199/18 (study #99550). In this study, rats were treated with H 199/18 by a single oral dose (gavage) at 0, 1000, and 2000 mg/kg (males) or 0, 900, and 1800 mg/kg (females). The maximum plasma levels of H 199/18 were ~64 and 119 µmol/l for 1000 and 2000 mg/kg males or 281 and 437 µmol/l for 900 and 1800 mg/kg females, respectively. AUC_{∞} values were 6670 and 16100 µmol.min/l for 1000 and 2000 mg/kg males or 49900 and 107000 µmol.min/l for 900 and 1800 mg/kg females, respectively. The percentage of protein binding was 68-83%.

Micronucleus Test of H 199/18 in Mice by Oral Gavage

(97484)

Testing Laboratory: Sponsor's laboratory

Dates Started and Completed: March 19, 1998 and May 28, 1998

Compliance With GLP and QAU Requirement: Sponsor included a statement of compliance with GLP regulation and a quality assurance statement.

Animals: Male CD-1 mice (31-40 g, 8-9 weeks old)

Drug Batch No.: 602/97, 102/98

Methods: To examine the potential mutagenic effects of H 199/18, micronucleus test was conducted using mouse bone marrow cells. A single dose of H 199/18 was given by oral gavage to male mice (7/group) at 0, 120, 620 and 1200 mg/kg. The dose selection was based on the results of dose ranging test. In this test, H 199/18 was tested orally at 1100, 1400, 1700 and 1900 mg/kg. There were two deaths (one male each at 1700 and 1900 mg/kg). The reduced motor activity was seen at all dose levels. One animal treated at 1400 mg/kg was immobile for more than 6 hours. Sponsor adequately chose the high dose of 1200 mg/kg in the current study. Mice were sacrificed 24 or 48 hours after dosing and bone marrow was collected. Vehicle and positive controls (cyclophosphamide) were also tested. The frequency of micronucleated polychromatic erythrocytes was then determined.

Results: Reduced motor activity was observed in all high dose animals. One high dose animal died within 24 hours after dosing. The reduced motor activity was also seen in a few low and mid dose animals. H 199/18 did not significantly increase the frequency of the micronucleated polychromatic erythrocytes as compared to the control. The positive control, however, significantly increased the frequency of the micronucleated polychromatic erythrocytes compared to the control.

In conclusion, H 199/18 was not mutagenic in this test system.

Addendum: A toxicokinetic study was conducted in mice to determine the plasma level of H 199/18 (study #99551). In this study, mice were treated with H 199/18 by a single oral dose (gavage) at 0, 620, and 1200 mg/kg. The maximum plasma levels of H 199/18 were ~240 and 482 µmol/l for 620 and 1200 mg/kg, respectively. $AUC_{0-\infty}$ values were 16700 and 53200 µmol.min/l for 620 and 1200 mg/kg, respectively. The percentage of protein binding was 74-86%.

LABELING:

The labeling is according to 21 CFR, Subpart B. The following revisions in the labeling are recommended.

WITHHOLD 3 PAGES

Draft

Labeling

SUMMARY AND EVALUATION:

H 199/18 (Esomeprazole magnesium) is a S-enantiomer of the racemic proton pump inhibitor, omeprazole, which inhibits H^+/K^--ATPase activity in the gastric parietal cells and thus blocks the final step of the gastric acid secretion. The pharmacological activity of H 199/18 was characterized in the *in vitro and in vivo* animal models and compared with omeprazole and H 199/19, the R-enantiomer. The results of the *in vitro* studies indicated that

H 199/18, H 199/19, and omeprazole inhibited histamine-induced acid formation in isolated gastric glands in rabbits with IC_{50} of 0.16, 0.27, and 0.086 mg/l, respectively. The results of the *in vivo* study indicated that H 199/18, H 199/19, and omeprazole inhibited gastric acid secretion with similar potency following both i.v. and oral administrations in rats.

In the present NDA, sponsor is seeking for approval to market Esomeprazole magnesium for treatment of gastroesophageal reflux disease (GERD) and erosive esophagitis. For symptom resolution and healing in patients with erosive esophagitis, the recommended dose is — mg once a day for 4-8 weeks. For prevention of the relapse and maintenance of symptom resolution of erosive esophagitis, the recommended dose is — mg once a day. For treatment of symptomatic GERD, the recommended dose is 20 mg once a day for 4 weeks. The dose of — mg/day represents a dose of — mg/kg/day if 50 kg body weight is assumed. In support of this NDA, the following studies were submitted: pharmacological studies, absorption, distribution, metabolism and excretion (ADME) studies in rats and dogs, toxicity studies: (1) acute toxicity studies in rats, (2) subchronic toxicity studies: 1- and 3-month oral toxicity studies in Wistar rats, 13-week oral toxicity study in Sprague-Dawley rats, and 3-month oral toxicity study in dogs, (3) reproductive toxicity studies: Segment II teratology studies in rats and rabbits, and (4) mutagenicity studies: Ames test, *in vitro* chromosome aberration tests using human peripheral blood lymphocytes, *in vivo* chromosome aberration test in rat bone marrow cells, and *in vivo* mouse micronucleus test.

The results of pharmacokinetic studies indicated that the maximum plasma concentrations of H199/18 and omeprazole were reached quickly within 3 minutes after intraduodenal administration in rats and within 18-26 minutes after gastric intubation in dogs. The intraduodenal bioavailability of H199/18 and omeprazole were 33% and 39%, respectively, in rats. The plasma levels of H199/18 and omeprazole declined quickly with terminal half life of 9-10 minutes in rats and 32-37 minutes in dogs. In humans, the plasma levels of H199/18 and omeprazole declined slower than those in rats and dogs with half lives of 1.0-1.2 hours in GERD patients. In both rats and dogs, very little (1.5-2%) racemization or inversion of H199/18 to H199/19 or vice versa occurs, suggesting that these enantiomers are resistant to inversion. Both H199/18 and omeprazole were excreted mainly in feces (57-61%) and in urine (30-31%).

In the acute toxicity studies in rats, the minimal lethal

oral dose of H 199/18 was identified at 990 mg/kg in males or 510 mg/kg in females. The minimal lethal oral dose of omeprazole sodium was 990 mg/kg. The minimal lethal i.v. dose of H 199/18 and omeprazole was 310 mg/kg and 240-310 mg/kg, respectively. The major treatment related clinical signs of toxicity in the acute toxicity studies were salivation, dyspnea, reduced motor activity, increased or decreased respiratory frequency, abdominal respiration, ataxia, tremor, convulsions, and cyanosis.

In the 1-month oral toxicity study in rats, H 199/18 was tested orally at 0, 15, 48, and 150 mg/kg/day in Wistar rats for 1 month. H 199/19 (15, 48, and 150 mg/kg/day) and omeprazole sodium and omeprazole (150 mg/kg/day) were also tested by oral gavage for 1 month. The major treatment related changes were decreased body weight gain (13-26%), increased salvation, and histopathological changes in the stomach (minimal foci of the chief cell eosinophilia in gastric mucosa) at 150 mg/kg/day of all four compounds (H199/18, H199/19, omeprazole sodium, and omeprazole). Decreased body weight gain was also noted in males treated with H 199/18 at 48 mg/kg/day (14.5%) and with H 199/19 at 48 mg/kg/day (8.6%). The stomach was the target organ of toxicity.

In the 3-month oral toxicity study in rats, H 199/18 was tested orally at 0, 14, 69, and 280 mg/kg/day in Wistar rats for 3 months. A separate group of rats received omeprazole at 140 mg/kg/day. The major treatment related changes were decreased body weight gain (16%, 13%, 28% in the low, mid, and high dose males, respectively) and histopathological changes (high dose) in the stomach (the chief cell eosinophilia at vacuolated glandular cells) and in the kidney (basophilic cortical tubules and inflammatory cell infiltration). Decreased body weight gain (18%) and histopathological changes in the stomach and kidney were also found in the omeprazole treated animals. The stomach and kidney were the target organs of toxicity.

In the 13-week oral toxicity study in rats, H 199/18 was tested orally at 0, 14, 69, and 280 mg/kg/day in Sprague-dawley rats for 13 weeks. A separate group of rats received omeprazole at 140 mg/kg/day. The major treatment related changes were mainly in the high dose group and omeprazole group. These included decreased hemoglobin, packed cell volume, corpuscular volume, increased platelet count, and histopathological changes in the stomach (the chief cell eosinophilia, acanthosis, and hyperkeratosis) and in the kidney (basophilic cortical tubules). There were no marked difference between groups of H 199/18 and omeprazole. The stomach and kidney were the target organs of

toxicity.

In the 3-month oral toxicity study in dogs, H 199/18 was tested orally at 0, 0.65, 5.5, and 28 mg/kg/day in dogs for 3 months. A separate group of dogs received omeprazole at 28 mg/kg/day. Histopathological changes were observed in the stomach and these included mucosal fibrosis, hyperplasia, chief cell atrophy, and focal necrosis in all treatment groups including omeprazole. There were no marked difference between groups of H 199/18 and omeprazole. The stomach was the target organ of toxicity.

The toxic profiles of H199/18 were characterized in rats and dogs. The stomach and kidney were the target organs of toxicity as evidenced by histopathological changes in the stomach (minimal foci of the chief cell eosinophilia, acanthosis, and hyperkeratorsis, mucosal fibrosis, hyperplasia, chief cell atrophy, and focal necrosis) and kidney (basophilic cortical tubules and inflammatory cell infiltration) in 1-month, 3-month oral toxicity studies in rats and 3-months oral toxicity study in dogs. Similar histopathological changes were observed following treatment with omeprazole in these studies.

In the Segment II teratological reproductive toxicity study in rats, H 199/18 was given to rats by oral gavage at 0, 14, 69, and 280 mg/kg/day during pregnant days 6 through 16. Maternal toxicity was evidenced by decreased body weight gain in the high (20%) dose group. There were no significant treatment related changes on number of corpora lutea, implantations, number of dead and live fetuses, body weight of the live fetuses, placental weight, and sex ratio. There were no treatment related changes in the major malformations. H 199/18 was not teratogenic in this study.

In the Segment II teratological reproductive toxicity study in rabbits, H 199/18 was given to pregnant rabbits by oral gavage at 0, 6.9, 28, and 86 mg/kg/day during pregnant days 6 through 16. Maternal toxicity was evidenced by decreased body weight gain in the mid (31%) and high (80%) dose groups. There were no significant treatment related changes on number of corpora lutea, implantations, number of dead and live fetuses, body weight of the live fetuses, placental weight, and sex ratio. There were no treatment related changes in the major malformations. H 199/18 was not teratogenic in this study.

Omeprazole was not teratogenic in Segment II oral teratological reproductive toxicity study in rats and rabbits.

However, in the Segment II teratological reproductive toxicity study in rabbits, omeprazole produced dose-related increases in embryo-lethality, fetal resorptions, and pregnancy disruptions in a dose range of 6.9 to 69.1 mg/kg/day. In rats, dose-related embryo/fetal toxicity and postnatal developmental toxicity were observed in offspring resulting from parents treated with omeprazole in a dose range of 13.8 to 138 mg/kg/day.

No carcinogenicity studies were conducted with H199/18. Sponsor included the findings of carcinogenicity studies with omeprazole. Treatment with omeprazole at oral doses of 1.7, 3.4, 13.8, 44, and 140.8 mg/kg/day produced gastric enterochromaffin-like (ECL) cell hyperplasia and carcinoids in both male and female rats in a dose dependent manner in two 2-year rat carcinogenicity studies. The results of a 78-week mouse carcinogenicity study of omeprazole showed no carcinogenic potential but this study was considered inconclusive and invalid by the Division as well as CDER Carcinogenicity Assessment Committee (CAC). Therefore, the findings of the 78-week mouse carcinogenicity study cannot be used for safety assessment.

[redacted]

H199/18 was negative when tested in Ames test, *in vivo* chromosome aberration test in rat bone marrow cells, and *in vivo* mouse micronucleus test. H199/18, however, was positive in two *in vitro* chromosome aberration tests using human peripheral blood lymphocytes. Omeprazole was also tested in one of these *in vitro* chromosome aberration tests using human peripheral blood lymphocytes and the result was positive. In addition, the clastogenic activity of omeprazole was also demonstrated in another *in vitro* human lymphocyte chromosomal aberration test (see amendment #185 dated February 13, 1992 in IND [redacted]. Omeprazole also displayed positive responses in the mouse micronucleus test and *in vivo* mouse bone marrow chromosomal aberration test.

In the present NDA, sponsor is seeking approval to market esomeprazole for treatment of gastroesophageal reflux disease (GERD) and erosive esophagitis and for prevention of the relapse and maintenance of symptom resolution of erosive esophagitis.

From a preclinical standpoint, approval of esomeprazole for short term use up to 8 weeks is recommended. However, due to genotoxic and tumorigenic potentials associated with esomeprazole and/or omeprazole, and lack of carcinogenicity study in second species (the mouse), approval of esomeprazole for long term use is not recommended from a preclinical standpoint. Relevant findings of the preclinical studies should be included in the labeling as recommended. Sponsor should be asked to revise the labeling as recommended.

RECOMMENDATION:

(1) From a preclinical standpoint, approval of esomeprazole for short term use up to 8 weeks is recommended, and approval of esomeprazole for long term use is not recommended.
(2) Sponsor should be asked to revise the labeling as recommended.

/S/ 8/8/00
Ke Zhang, Ph.D. Date
Pharmacologist, HFD-180

Comments: (1) Concur. (2) Sponsor conducted the recommended studies as provided in "FDA's Policy Statement for the Development of New Stereoisomeric Drugs, F.R. Notice 57(102), May 27, 1992". 3) If Supervisory Pharmacologist memo on labeling will follow.

/S/ 8/23/00
Jasti B. Choudary, B.V.Sc., Ph.D. Date
Supervisory pharmacologist, HFD-180

cc:
NDA
HFD-180
HFD-181/CSO
HFD-180/Dr. Choudary
HFD-180/Dr. Zhang
HFD-345/Dr. Viswanathan

R/D Init.: J. Choudary 8/1/00

KZ/Deg: 8/8/00