Mutagenicity:

B.4.1 Evaluation of AD 32 in the Ames Assay with E. Coli (sic). 1996. Study 3-D22. Vol 1.23, page 55.

Drug	AD 32, (OLI-222) Lot 002.
Bacteria	S. typhimurium TA98, TA100, TA1535, TA1537 all with rfa mutation
	E. coli WP2 uvr A, WP2 uvr A with pKM101.
Solvent	DMSO
Dose	in a range finding study and in confirmatory assay 0.050 mg/plate to 5 mg/plate.
Positive Cntl.	2-aminoanthracene with S9
	2-nitrofluorene (TA98)
	Sodium azide (TA100 and TA1535)
	9-aminoacridine (TA1537)
	ENNG (WP2 uvr A both strains)
Metabolic Act.	S9 from Aroclor 1254 induced Sprague-Dawley rats.

This study was done by signed the GLP statement.

I do not consider Aroclor 1254 the best inducer of S9 for this experiment. It primarily induces CYP 1A1 in the rat. Human liver does not express this isoform in large concentrations. Nevertheless, in this experiment this problem is irrelevant because AD 32 was strongly mutagenic with and without S9. The following table shows that AD 32 caused an increase in the mean number of revertants per plate in two Salmonella strains, TA98, TA100, and one E. coli strain, WP2 uvrA with pKM101. All three of these bacterial strains contained the pKM101 plasmid. This plasmid increases the sensitivity of the bacteria to mutations involving error prone repair. AD 32 did not cause and increase in the number of mutations in the three strains that did not contain this plasmid. This implies that mutations caused by AD 32 involve repair mechanisms. Metabolic activation increased the frequency of mutations in strain TA98, and perhaps TA100. Activation seemed to decrease the frequency of mutations in the E. coli strain.

	S9	TA98 pKM101	TA100 pKM101	TA1535	TA1537	WP2uvr A	WP2uvr A pKM101
Negative Control	(-)	16	110	22	13	52	80
	(+)	23	121	17	11	74	75
Positive Control	(-)	737	660	347	473	195	355
	(+)	2271	2874	435	458	450	1624
AD 32 mg/plate							
5	(-)	564	346	16	11	54	161
1.5	(-)	386	288	20	11	49	133
0.5	(-)	299	273	18	10	47	107
0.15	(-)	131	1899	25	13	52	85
0.05	(-)	39	141	22	11	55	61
5	(+)	857	356	14	13	66	117
1.5	(+)	785	338	15	14	62	104
0.5	(+)	315	297	16	20	64	103
0.15	(+)	154	288	18	19	72	101
0.05	(+)	78	227	17	17	64	90

The compound precipitated at 1.5 and 5 mg/plate. Thus, the dose response may diminish at these concentrations or above. The lowest concentration tested, 0.05 mg/plate, is somewhat higher than the usual low concentration in an Ames assay. This low concentration shows a positive response in TA98 and TA100. This experiment would be more informative if the investigators had extended their concentration series a log lower.

B.4.2. Evaluation of AD 32 in the *in vitro* chromosomal aberration assay. Study 3-D23. 1997, Vol 1.23, p 98.

Drug	AD 32, (OLI-222) Lot 002.
Cells	Chinese hamster ovary (CHO).
Solvent	DMSO
Dose	0.075 to 75 µg/plate activated range finding assay 0.25 to 2500 µg/plate non-activated range finding assay 0.25 to 2.5 µg/plate non-activated confirmatory assay
Exposure time	3 hr
growth time	18 hr
Positive Cntl.	Mitomycin C without S9 Cyclophosphamide with S9
Metabolic Act.	S9 from Aroclor 1254 induced Sprague-Dawley rats.

This study was done by signed the GLP statement.

The following table shows that AD 32 causes even more chromosomal damage than the positive control, mitomycin C, without metabolic activation. The addition of metabolic activation does not greatly increase this damage. The concentrations that cause aberrations (ABS) are relatively low, 0.75 µg/ml or 1 µM. The authors of this study say that the compound

caused a variety of complex damage including interstitial deletion, triradial, quadriradial, dicentric ring, and complex rearrangement. AD 32 is clastogenic to mammalian cells *in vitro*.

			Metabolic Activation Gaps Excluded			Metabolic Activation Gaps Excluded		
	Dose	No. Cells Analyzed	Total No. ABS	Mean ABS/cell	% Cells With ABS	Total No. ABS	Mean ABS/cell	% Cells With ABS
	µg/ml							
Positive Control								
Cyclophosphamide	25	25	85	3.4	92			
		25	82	3.28	92			
Mytomycin C		25				53	2.12	76
		25				40	1.6	60
Negative Control								
DMSO	1%	100	2	0.02	2	2	0.02	2
		100	1	0.01	1	1	0.01	1
AD 32	0.25	100	2	0.02	2	3	0.03	3
		100	1	0.01	1	3	0.03	3
	0.75	100	6	0.06	4	45	0.45	27
		100	11	0.11	10	27	0.27	20
	2.5	25	161	6.44	96	173	6.92	96
		25	107	4.28	76	158	6.32	100

Mutagenicity Summary

AD 32 caused increases in the number of bacterial revertants as high as 17 fold above control in two strains of Salmonella and one strain of E. coli without S9 activation. With S9, AD 32 caused increases in the number of revertants as high as 53 fold. The strains that were sensitive all contained the pKM101 plasmid suggesting that AD 32 causes mutations involving error prone repair. In *in vitro* chromosomal aberration assays, AD 32 caused even more chromosomal damage than the positive control, mitomycin C, without S9. At the higher concentrations the incidence of damage was more than 300 fold greater than control. Adding S9 did not significantly increase this damage. Clearly, AD 32 is mutagenic.

Reproductive toxicity

D. 1. Intravenous developmental toxicity study of AD 32 in Rats. Study 2901-001. Submission BP, received March 26, 1998.

Animals	Crl:CD Sprague-Dawley female Rats
Drug	AD32 Sterile Liquid, lot BVL 515-44-0003
Vehicle	NCI Diluent 12 in 0.9% NaCl
Doses	0, 36, 72, 144 mg/m^2 (given as 0, 6, 12 and 24 mg/kg)
Concentrations	0, 6, 12 and 24 mg/ml
Route	IV
Schedule	daily on gestation days 7 through 17
N	25 per dose group, plus a satellite group with 2 in the control and 6 in the test.
Observations	
Clin. Obs.	Daily
Food Cons.	d0, 7, 10, 12, 15, 18, and 20
Body Wt.	daily
Blood	d7 before and after dosing in 3 dosed satellite rats these rats were killed 4 hr after dosing. D17 before and after dosing in 3 dosed satellite rats and all fetuses these rats were killed 4 hr after dosing.
Necropsy	D20

~ half of the fetuses in each litter were examined for soft tissue alterations
~ half of the fetuses in each litter were examined for skeletal alterations

did this study for Anthra
Pharmaceuticals. signed the GLP statement.

Results

Mortality	No deaths, abortions or premature deliveries occurred during the study.
Clinical Obs.	None associated with AD 32 dosing
Body Wt.	Dose dependent decrease in mid and high dose group animals independent of fetal resorption. (to 11% decrease relative to control in HD at day 20)
Food Cons.	Consistent with body weight decrease.

Caesarean Section Results

	Control			6 mg/kg			12 mg/kg			24 mg/kg		
	N	mean/litter	SD	N	mean/litter	SD	N	mean/litter	SD	N	mean/litter	SD
Number of Rats tested	25			25			25			25		
Corpora Lutea		16.2	2		16	1.9		17	3.1		15.9	2.4
Implantations		15.1	1.1		14.9	1.2		15.4	2.5		14.7	1.8
Litter Sizes		14.5	1.5		14.5	1.3		15.2	2.7		9.8*	6.3
Live Fetuses	333	14.5	1.5	333	14.5	1.3	364	15.2	2.7	244	9.8*	6.3
Dead Fetuses	0			0			0			0		
Resorptions		0.6	0.8		0.4	0.6		0.2	0.5		4.9	6.2
Early Resorptions	15	0.6	0.8	9	0.4	0.6	5	0.2	0.5	111	4.4	6.2
Late Resorptions	0			0			0			12	0.5	1.2
		Percentage			Percentage			Percentage			Percentage	
Pregnant	23	92		23	92		24	96		25	100	
Dams with any resorptions	11	47.8		8	34.8		4	16.7		15	60	
Dams with all conseptuses resorbed	0			0			0			6	24**	
Dams with viable fetuses	23	100		23	100		24	100		19	76**	
Placentae appeared normal	23	100		23	100		24	100		19	100	

* $p \leq 0.05$

** $p \leq 0.01$

Incidence of any Fetal Alterations

	Control		6 mg/kg		12 mg/kg		24 mg/kg	
Litters Evaluated	23		23		24		19	
Fetuses Evaluated	333		333		364		244	
Live	333		333		364		244	
Litters with fetuses with any alteration N and %	8	34.8	10	43.5	16*	66.7	18**	94.7
Fetuses with any alteration N and %	10	3	12	3.6	58	15.9	113**	46.3
% fetuses with any alteration per liter, mean and SD	2.8	4.2	3.5	4.5	15.7**	17.2	52.5**	37.7

* $p \leq 0.05$

** $p \leq 0.01$

Malformations

The 12 and 24 mg/kg/d dose groups had dose-dependent increases in litter and fetal incidences of depressed eye bulges, short snout and tail malformations. The litter and fetal incidences of depressed eye bulges and short snout were significant in the 24 mg/kg/d group. Additional increased incidences of external malformations in the HD group included head malformations (meningocele, cleft snout with associated cleft palate, protrusion of the tongue, dilation of the ventricles of the brain), edematous body and umbilical hernia. The following table shows the litter and fetal incidence of some of these malformations that reached statistical significance (** $p \leq 0.01$).

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY)a		control	6	12	24
LITTERS EVALUATED	N	23	23	24	19
FETUSES EVALUATED	N	161	160	177	118
LIVE	N	161	160	177	118
PALATE: CLEFT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (1.7)**
EYES: MICROPHTHALMIA					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	4 (16.7)	11 (57.9)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	10 (5.6)	33 (28)
BRAIN: LATERAL VENTRICLES, MODERATE DILATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	1 (0.6)	6 (5.1)**
BRAIN: LATERAL VENTRICLES, MARKED DILATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	4 (16.7)	9 (47.4)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	5 (2.8)	20 (16.9)**
BRAIN: THIRD VENTRICLE, MODERATE DILATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	7 (36.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	2 (1.1)	12 (10.2)**
BRAIN: THIRD VENTRICLE, MARKED DILATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (21)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	7 (5.9)**
BRAIN: THIRD VENTRICLE, SLIGHT DILATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	4 (16.7)	2 (10.5)*
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	4 (2.2)	7 (4.2)**
BRAIN: LATERAL VENTRICLES, EXTREME DILATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)

Skeletal Alterations

Skeletal alterations were numerous and dose related. In the skull the incidence of small eye socket was associated with microphthalmia seen above. Small nasal turbanates, premaxillae and maxillae or incompletely ossified sphenoid was associated with short snout. In the cervical vertebrae, the incidences of fused arches or small arches or both increased in the mid and high dose groups. The incidence of fused ribs was significant in the mid and high dose groups. The high dose group had additional rib malformations including segmentation or absence of the rib or the rib base, reduced rib number and broadening of the ribs. In this group there was also an increase in the incidence of "scrambling" of the thoracic vertebral elements (bifid, not ossified, unilaterally ossified or irregularly shaped or fused centra, fused arches, small arches, or hemivertebra). In the lumbar region the high dose group showed an increase in bifid centra.

Variations of the cervical vertebrae included the presence of cervical ribs at the 7th cervical vertebra in the mid and high dose groups. Delayed and incomplete ossification of the sternum was significant in the high dose group. The following table shows the litter and fetal incidence of all the alterations and variations that reached statistical significance and some that did not.

PROTOCOL 2901-001: INTRAVENOUS DEVELOPMENTAL TOXICITY STUDY OF AD 32 IN RATS
TABLE 11 (PAGE 1I: FETAL SKELETAL ALTERATIONS - SUMMARY
(See footnotes on the last page of this table.)

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY)		control	6	12	24
LITTERS EVALUATED	N	23	23	24	19
FETUSES EVALUATED	N	172	173	187	126
LIVE	N	172	173	187	126
SKULL: NASALS, SMALL					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	4 (21.0)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	1 (0.5)	9 (7.1)**
SKULL: MAXILLAE, SMALL					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	4 (21.0)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	1 (0.5)	9 (7.1)**
SKULL: PREMAXILLAE, SMALL					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	4 (21.0)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	1 (0.5)	9 (7.1)**
SKULL: EYE SOCKET, SMALL					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	2 (8.3)	12 (63.2)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	4(2.1)	45 (35.7)**
SKULL: SPHENOID, INCOMPLETELY OSSIFIED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.2)**
SKULL: ANTERIOR FONTANELLE, LARGE					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
SKULL: PALATE, INCOMPLETELY OSSIFIED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
CERVICAL VERTEBRAE: CERVICAL RIB PRESENT AT 7TH CERVICAL VERTEBRA					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	12 (50.0) **	12 (63.2)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	23 (12.3)**	22 (17.5)**
CERVICAL VERTEBRAE: ARCHES, FUSED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	9 (47.4)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	1 (0.5)	19 (15.1)**
CERVICAL VERTEBRAE: ARCH, SMALL					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	1 (4.2)	9 (47.4)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	1 (0.5)	14 (11.1)
CERVICAL VERTEBRAE: ARCH, NOT OSSIFIED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	7 (36.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	16 (12.7)**
CERVICAL VERTEBRAE: 6 PRESENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
CERVICAL VERTEBRAE: IRREGULARLY SHAPED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY)a		control	6	12	24
LITTERS EVALUATED	N	23	23	24	19
FETUSES EVALUATED	N	172	173	187	126
LIVE	N	172	173	187	126
THORACIC VERTEBRAE: CENTRUM, BIFID					
	LITTER INCIDENCE	2 (8.7)	2 (8.7)	6 (25)	12 (63.2)**
	FETAL INCIDENCE	2 (1.2)	2 (1.2)	8 (4.3)	22 (17.5)**
THORACIC VERTEBRAE: CENTRUM, UNILATERAL OSSIFICATION					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	7 (36.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	14 (11.1)**
THORACIC VERTEBRAE: CENTRA, FUSED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (2.4)**
THORACIC VERTEBRAE: ARCHES, FUSED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	5 (26.3)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	6 (4.8)**
THORACIC VERTEBRAE: ARCH, SMALL					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.2)**
THORACIC VERTEBRAE: CENTRUM, NOT OSSIFIED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.2)**
THORACIC VERTEBRAE: HEMIVERTEBRA					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (2.4)**
THORACIC VERTEBRAE: 11 PRESENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
THORACIC VERTEBRAE: 12 PRESENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (10.5)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (2.4)**
LUMBAR VERTEBRAE: CENTRUM, BIFID					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.2)**
RIBS: FUSED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	7 (36.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	16 (12.7)**
RIBS: TWO SEGMENTS					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (10.5)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (1.6)**
RIBS: ABSENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
RIBS: 11 PRESENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (1.6)**
RIBS: BROAD					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.2)**

DOSAGE GROUP		I	II	III	IV
DOSAGE (MG/KG/DAY)a		control	6	12	24
LITTERS EVALUATED	N	23	23	24	19
FETUSES EVALUATED	N	172	173	187	126
LIVE	N	172	173	187	126
RIBS: BASE ABSENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (15.8)**
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.2)**
RIBS: 12 PRESENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (10.5)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	3 (2.4)**
RIBS: 10 PRESENT					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
MANUBRIUM: MISALIGNED					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (5.3)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.8)
STERNAL CENTRA SUMMARIZATION (Includes incompletely ossified, not ossified, fused and asymmetric sternal centra)					
	LITTER INCIDENCE	5 (21.7)	2 (8.7)	6 (25.0)	12 (63.2)**
	FETAL INCIDENCE	5 (2.9)	3 (1.7)	12 (6.4)	26 (20.6)**
STERNAL CENTRA: 1ST, NOT OSSIFIED					
	LITTER INCIDENCE	2 (8.7)	1 (4.3)	3 (12.5)	8 (42.1)**
	FATAL INCIDENCE	2 (1.2)	1 (0.6)	6 (3.2)	16 (12.7)**
STERNAL CENTRA: 1ST, INCOMPLETELY OSSIFIED					
	LITTER INCIDENCE	3 (13.0)	2 (8.7)	5 (20.8)	9 (47.4)**
	FETAL INCIDENCE	3 (1.7)	2 (1.2)	6 (3.2)	9 (7.1)**
STERNAL CENTRA: ASYMMETRIC					
	LITTER INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (10.5)
	FETAL INCIDENCE	0 (0.0)	0 (0.0)	0 (0.0)	2 (1.6)**

In conclusion, the maternal NOAEL of AD 32 is less than 6 mg/kg/d since rats in all dose groups lost weight. Thus all doses in this experiment caused maternal toxicity. Likewise, 6 mg/kg/d was not a fetal NOAEL, all dose levels caused fetal body weight reduction. Higher doses, 12 and 24 mg/kg/d, caused embryo-fetal death and numerous fetal malformations and variations, particularly in the skeletons of the fetal mice.

Pharmacokinetics and Toxicokinetics:

C.2. M Israel and VK Khetarpal, 1991. Comparative tissue distribution and metabolism studies with Dox and AD 32 in normal and tumor bearing mice. Vol. 1.24, page 300.

did this impressively large and comprehensive study to compare the metabolism and distribution of AD 32 and Dox. They wanted to determine why AD 32 appears to have a significantly better therapeutic index in certain mouse tumor models than Dox. AD 32 causes less acute toxicity and less cumulative cardiac toxicity. AD 32 is more lipophilic than Dox. This may explain why AD 32 is cleared from the blood into tissue somewhat more quickly. In cells AD 32 appears to accumulate in the cytoplasm. The hypothesis underlying these studies was that the increased effectiveness of AD 32 resulted from this greater and more rapid

accumulation in cells followed by its enzymatic conversion to Dox. That is, is AD 32 a well distributed Dox prodrug?

The authors gave male A/JAX mice 50 mg/kg AD 32 or 4 mg/kg Dox IV tail vein. They killed five animals at each time point (15 and 30 min, 1, 2, 4, 6, 8, 12, 16, and 24 hr after treatment) and took tissue samples. They homogenized and extracted the tissue samples and quantitated the anthracycline concentrations by with fluorescence detection. In similar experiments they examined tissues from mice with tumor implants and mice given four daily doses of anthracycline.

After a single dose, AD 32 disappeared rapidly from the serum and from most tissues. This was due to rapid hydrolysis to AD 41. AD 92 was also detected in all tissues, but the highest concentrations occurred in the small intestine (with contents). The authors detected and characterized two previously unrecognized aglycone metabolites, AD 151 in most tissues, and AD 112 in the liver and kidney. Dox was detected as a minor metabolite.

A single dose of Dox was also rapidly cleared from serum. Nevertheless, in this case the parent compound was little metabolized. AMNOL (dihydroadriamycin), adriamycinone, AD151 and AD 112 were minor metabolites. Dox tissue concentrations were 3 to 8 times less in AD 32 treated mice than in Dox treated animals.

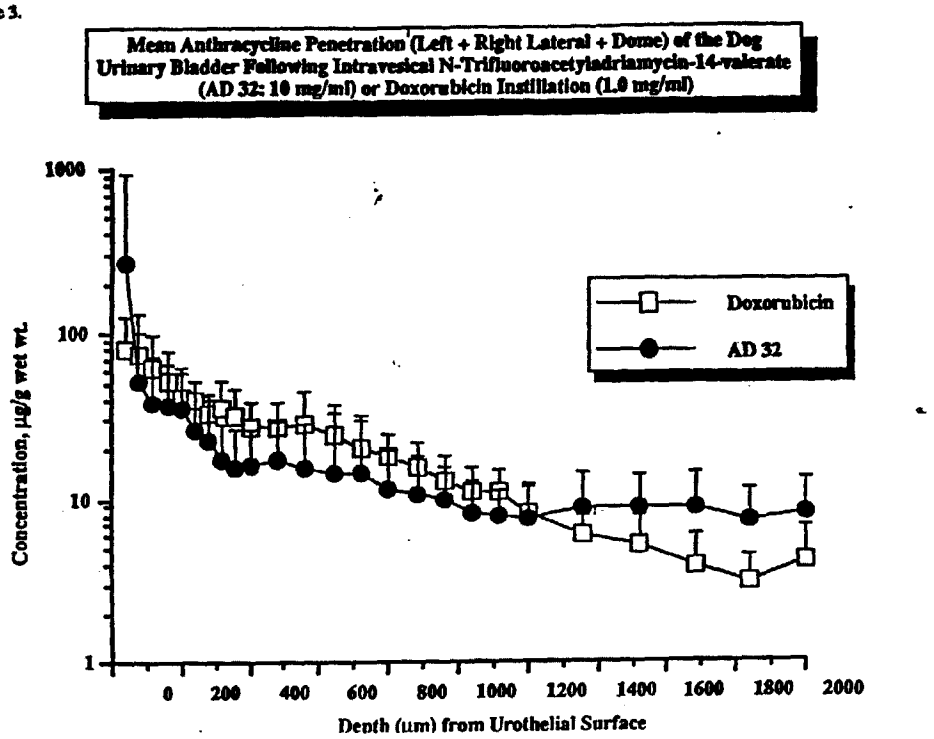
Daily treatment for four days with AD 32 and Dox caused accumulation of Dox in tissues, but Dox concentrations in AD 32 treated mice were still 3-8 fold lower than those in Dox treated mice. In tumor bearing mice, Dox concentrations were also lower in AD 32 treated animals than in Dox treated animals. Again in this study as in others, AD 32 was more effective against tumor implants than AD 32 at toxicologically equivalent doses. These results suggest that AD 32 is not a simple pro-drug of Dox and that the differences in its cytotoxicity, and hence its anti-tumor activity, are related to the presence of the N-trifluoroacetyl group.

C.4. TW Sweatman. N-trifluoroacetyladriamycin-14-valerate (AD32): Drug penetration of the dog urinary bladder following intravesical instillation, 1992. Volume 1.12, page 086. This report is identical to study A.2. Vol. 12, p 218.

Sweatman instilled AD 32 (400 mg in 40 ml 25% NCI Diluent 12 plus 75% saline) or Dox (40 mg in 40 ml saline) into the urinary bladders of conscious sedated dogs. Fifteen minutes before the end of the installation he anesthetized the dogs and surgically exposed the urinary bladder. He removed the drug and accumulated urine and then excised the bladder. The bladder was immediately frozen on an aluminum block. Sequential 10 micron sections, parallel to the epithelial surface, were cut from three 2 cm X 2 cm squares of tissue, one each from the left and right lateral surfaces and one from the bladder dome. Three dogs were used for each chemical. The anthracyclines were then extracted from groups of the sections. The concentration of each anthracycline was determined in the samples by

The following graph shows the results of this experiment. The text does not clearly state that the concentrations in the graph are those of AD 32 or of AD 32 and its metabolites. My impression is that it is the latter. My comments are based on that assumption. The text does imply that the technique quantified AD 41 in the samples. Dox had no metabolites under these conditions.

Figure 3.



The urinary bladder of the dog is morphologically similar to that of humans, but it is only half as thick. Nevertheless, diffusion distance is probably more a function of morphology than of thickness. The concentration of both chemicals falls steeply between the luminal surface and approximately 200 microns. T1 tumors are located in the region of the lamina propria, or about 400 microns deep in the dog. At this level the mean concentration of AD 32 was 16000 ng/g wet weight or approximately 22.1 µM assuming a specific gravity of 1. Deeper within the superficial muscle layers of the bladder wall at the level of T2 tumors (~1600 microns) the concentration of anthracycline remained essentially constant across a wide band of tissue.

Overall Summary:

AD 32 is a synthetic analog of adriamycin (Dox). The addition of a valerate ester to the 14-carbon significantly increases the lipid solubility of this compound over that of adriamycin. Thus, AD 32 crosses the cell membrane rapidly and unlike Dox, accumulates in the cytoplasm. AD 32 does not appear to intercalate with DNA. AD 32 is extensively metabolized and cleared rapidly when given to test animals or humans IV. AD 32 is fluorescent. This property allows small concentrations of the compound to be determined.

Four daily doses of 12 mg/m^2 (given as 4 mg/kg) of Dox and 120 to 180 mg/m^2 (40 to 60 mg/kg) AD 32 in tumor bearing mice are roughly equipotent against a range of tumors. At 12 mg/m^2 Dox is somewhat more toxic than equivalent doses of AD 32, thus AD 32 appears to have a better therapeutic index than Dox. This observation and the fact that AD 32 appears to be effective against some tumor implants resistant to Dox seems to have driven much of the research into the pharmacology of this compound.

AD 32 is not effective against intracerebral tumor implants, implying that it does not cross the BBB. *In vivo*, AD 32 is an even more potent immunosuppressant than Dox. This immunosuppression is probably the simple result of myelotoxicity. Like that of Dox, the myelotoxicity associated with AD 32 is very rapid; nadir occurs by day two of dosing.

The major metabolite of AD 32 is AD 41 in all tissues studied. Cytochrome P450 oxidizes AD 32 to more polar compounds only in very limited concentrations in the liver. Unlike Dox, AD 32 does not inhibit DNAse I.

DNA added to AD 32 preparations, did not quench the fluorescence of AD 32. This suggests that the aromatic region of AD 32 does not interact directly with the DNA.

A single IP dose of 54 mg/kg (324 mg/m^2) caused no adverse effects in female albino rats. A dose of 106 mg/kg (636 mg/m^2) caused only mild toxicity including transient anorexia, decrease in segmented neutrophils.

When investigators instilled doses of AD-32 (54 mg/kg, 34 mg/ml) in the urinary bladder of anesthetized rats they found only mild signs of irritation. These signs were also seen in controls. The bioavailability of AD-32 after instillation in the urinary bladder of rats is < 1%. Recovery of the dose from the bladder after instillation was > 95% after 2 hr.

AD 32 did not cause corneal opacity, iritis or any unusual ocular toxicity when instilled in the eyes of rabbits. It caused only mild irritation probably associated with the vehicle. Likewise, AD 32 caused no unusual local skin irritation. AD 32 does not appear to cause the profound cumulative cardiac toxicity that Dox causes in rabbits.

The LD_{10} for a single dose of AD 32, IV, is 46 mg/kg (95% CL 35 to 56 mg/kg) in rabbits. Single doses to 636 mg/m^2 (given as 106 mg/kg), IP, caused only minor toxicity in rats when the dose was removed after 4 hours. Single doses to 53 mg/kg instilled in the urinary bladder of female rats for 2 hours caused no serious toxicity and no detectable microscopic damage to the bladder. Single intrathoracic doses of AD 32 of 600 mg/m^2 (given as 100 mg/kg) killed 6 of 10 female rats but an identical volume of vehicle control killed 4 of 10. A single intrathoracic dose of 30 mg/kg killed one of four beagle dogs. In both the intravesical and intrathoracic studies the vehicle, NCI Diluent 12, was dose limiting.

Three or six doses of AD 32 instilled in the urinary bladder of rats to 324 mg/m^2, seven days apart, caused only limited toxicity. The trauma and damage in the peritoneum, bladder and urethra were probably caused by the procedure. Six intravesical doses of AD 32 in the urinary bladder of male dogs to 400 mg caused neovasularization and the accumulation of pigment laden macrophages in the bladder epithelium. Treatment also caused atrophy and inflammation of the prostate, and testicular degeneration. Some of this damage remained after 35 days of recovery. Unfortunately the experiment did not include sufficient information about the vehicle controls, so I cannot distinguish AD 32 toxicity from vehicle toxicity (NCI Diluent 12).

Three IP doses to 636 mg/m^2 given 14 days apart to rats caused no mortality and only a 33% decrease in WBC. The doses in this study were among the highest systemic doses given to rats or dogs. The study suggests that myelosuppression is dose limiting, but the high dose was relatively low. Three IP doses to 600 mg/m^2 given 21 days apart to rats also caused no mortality. Nevertheless, this longer schedule did cause edema and degeneration of the sciatic nerve that increased in intensity with dose. This neuropathy is possibly associated with a

diminished blood-neural-barrier in the lumbosacral area of rats. The significance of this toxicity to humans is questionable. In this study, IP dosing caused damage to the testes even at 480 mg/m^2.

In beagle dogs, three IP doses to 400 mg/m^2 given 21 days apart caused only mild degeneration in the seminiferous tubules and some degeneration in the liver. These toxicities are not unusual for an anthracycline. The dose approximates the proposed human dose which is relatively low.

Repeated intrathoracic dosing in rats to 600 mg/m^2 caused the death of 20 of 20 animals. Half this dose was lethal to 14 of 20 animals. In this experiment, vehicle at the volume given to high dose animals caused the death of four of 20 animals. Death appeared to be caused by pulmonary effusion. In surviving animals dosing caused prolonged lymphopenia, adhesions, pericarditis, fibrinous pleuritis and gross changes in the liver and spleen. Three intrathoracic doses of 400 mg/m^2 given 21 days apart killed two of four dogs. Surviving animals suffered weight loss, an inflammatory response after dosing. Local toxicities included thoracic effusions, fibrin deposition and adhesions. Treatment with AD 32 by this route is probably too toxic to be a practical therapy.

Long term IV dosing with AD 32 appears to cause a different spectrum of toxicities than Dox. A total dose of 60.7 mg (0.11 mmoles) of Dox over 12 weeks caused cardiac edema ventricular dilation, myocytolysis, fibrosis and atrophy in rabbits. This cumulative dose of Dox also caused direct damage to the glomerulus in the kidneys, centrolobular necrosis in the liver and aspermia and testicular degeneration. A dose cumulative dose 492.1 mg of AD 32 (0.68 mmoles) over the same period caused only moderate cardiac edema and vacuolization in rabbits. In this study AD 32 also caused enlargement of the spleen, bone marrow hypoplasia and fatty changes in the liver. The splenic enlargement was associated with sinusoids filled with erythrocytes and granulocytes at various stages of maturity.

In humans, the dose limiting toxicity for IV AD 32 is myelosuppression. When the drug is instilled in the urinary bladder of humans, irritation from the vehicle (ethanol) is dose limiting at 900 mg.

AD 32 caused increases in the number of bacterial revertants as high as 17 fold above control in two strains of Salmonella and one strain of E. coli without S9 activation. With S9, AD 32 caused increases in the number of revertants as high as 53 fold. The strains that were sensitive all contained the pKM101 plasmid suggesting that AD 32 causes mutations involving error prone repair. In *in vitro* chromosomal aberration assays, AD 32 caused even more chromosomal damage than the positive control, mitomycin C, without S9. At the higher concentrations the incidence of damage was more than 300 fold greater than control. Adding S9 did not significantly increase this damage. Clearly, AD 32 is mutagenic.

In a study of reproductive toxicity, AD 32 caused significant decreases in litter size and early resorption (7 fold increase) at doses that only caused a 10% weight reduction in dams (72 mg/m^2 or approximately one sixth the proposed clinical dose). A dose of 18 mg/m^2 also caused some weight loss in the dams, so it was not a NOAEL. Doses of 36 and 72 mg/m^2 caused significant dose dependent fetal malformations and variations. These included, but were not limited to, microphthalmia, short snout (with attendant skeletal alterations), meningocele, cleft palate, protrusion of the tongue, dilation of the ventricles of the brain, fused ribs, alterations of the vertebra, and delayed or incomplete ossification. AD 32 causes embryo and fetal death and is teratogenic.

In monkeys the elimination of AD 32 from serum appears biphasic. The half-lives are 7 minutes and 3.8 hours. This compares to half-lives of 2.7 minutes and 12.5 minutes for Dox. After 24 hours, 30.4% of a given dose was eliminated in the bile primarily as AD 41 and AD 92. Monkeys eliminated 11% of a given dose in the urine. These results imply that AD 32

metabolites maintain relatively long residence in the body, and particularly the hepatobiliary system.

AD 32 is not a simple prodrug of Dox. Its major metabolite AD 41, is cytotoxic and has considerable anti-tumor activity. In humans, a single dose of AD 32 is rapidly cleared from the serum and most of it is rapidly converted to AD 41 and AD 92. Cytochrome P540 dependent metabolism is not a major factor in the elimination of AD 32. Urinary excretion of anthracyclines is between 4 and 9% of the total dose. As with most species studied, humans eliminate most of a given dose in the bile.

The systemic MTD of AD 32 in humans is approximately 900 mg/m^2. Most clinical protocols used a dose of 600 mg/m^2. The proposed clinical dose in this NDA is significantly less than these doses on a mg/m^2 basis. Thus, even if the entire dose entered the peritoneum by accident the procedure should not cause unmanageable toxicity.

Recommendation:

None of the preclinical information submitted in this NDA precludes approval of AD 32 for the proposed indication. These studies may proceed based on previous clinical and preclinical data.

NDA Status:

The oncology advisory committee voted not to approve this NDA because the clinical trials failed to establish efficacy.

Labeling Review:

The proposed label conforms to CFR 201.5.7. Editing will be needed should this drug be approved.

/S/

W. David McGuinn, Jr., Ph. D., D.A.B.T.

June 22, 1998

cc: Original IND
/HFD-150
/P Andrews
/A Staten
/WD McGuinn

6/23/98

NDA 20-671

Account of Histopathology samples

Study		3-E64	048-004	AD332RAT CHSTX4
Species		Dogs	Rat	Rat
Adrenals				X
	Cortex	X		
	Medulla	X		
Aorta		X		X
Bone				
Bone Marrow smear		X	X	
Brain		X		X
Brain Stem		X		
Cecum		X		
Cerebellum		X		
Cerebrum		X		
Cervix				
Chest wall				
	Right			X
	left			X
Colon		X		X
Diaphragm				X
Duodenum		X		X
Epididymis		X		X
Esophagus		X		X
Eye		X		X
Falopian tube				
Femur				
Foot			X	
Gall bladder		X		
Hind Limb			X	
Heart		X	X	X
Harderian gland				
Ileum		X		
Injection site				
Jejunum		X		X
Kidneys		X	X	X
Lachrymal gland				
Large Intestine			X	
lymph node		X	X	
Liver		X	X	X
Lungs		X	X	
Apical & diaphragmatic lobes right				X
Apical & diaphragmatic lobes right				X
Macroscopic lesions				
Mammary Gland		X		
Mesenteric lymph glands		X		
Ovaries			X	X
Pancreas		X		

	Histo		
Parathyroid	X		
Paricardium			X
Peripheral nerve	X		
Pituitary	X		
Prostate	X		
Rectum	X		
Salivary gland			X
Sciatic nerve		X	
Seminal vesicle			
Skeletal muscle	X		X
Skin		X	
Small Intestine	X		
Spinal cord	X	X	
Spleen	X		
Sternum	X		
Stomach	X		X
Testes	X	X	X
Thymus	X		X
Thyroid	X		
Tongue			X
Trachea	X		
Urinary bladder	X		X
Uterus			X
Vagina	X		
Injection Site	X	X	
tissue masses			
gross lesions			

Comprehensive List of studies submitted to NDA 20-894

See the review of this NDA for a list of the studies among these reviewed for the determination of approval.

APPENDIX A Individual Nonclinical Pharmacology Study Reports

A.1 Report NSC 246131. Adriamycin cardiotoxicity. A comparison with adriamycin, AD-32 N-trifluoroacetyladriamycin-14-valerate, 1979. SUBMITTED TO IND

A.2 Report. Sweatman TW. N-trifluoroacetyladriamycin-14-valerate (AD 32): Drug penetration of the dog urinary bladder following intravesical instillation, 1992. Vol 1.12, p 218

A.3 Report. Swaminathan, S. Cytotoxicity of AD 32 in human bladder cancer cell lines. 1997. Vol. 1.12, p 228

APPENDIX B Individual Toxicology Study Reports

B.1 Acute Toxicity Studies

B.1.1 Report WIL-178001. Acute study in female albino rats with AD 32 administered by urinary bladder catheterization, 1991. SUBMITTED TO IND

B.1.2 Report WIL-178002. Acute intraperitoneal toxicity study in female albino rats with AD 32, 1991. SUBMITTED TO IND

B.1.3 Report AD32RATCHSTX1. A single dose toxicity study of AD 32 injected directly into the thoracic cavity of rats, 1996.SUBMITTED TO IND

B.1.4 Report AD32CHSTX1. A single dose toxicity study of AD 32 injected directly into the thoracic cavity of beagle dogs, 1995. SUBMITTED TO IND

B.1.5 Report AD32PROSX1. A single dose toxicity study of AD 32 injected directly into the prostate of beagle dogs, 1995. SUBMITTED TO IND

B.2 Repeat-Dose Toxicity Studies

B.2.1 Report WIL-178004. Repeated exposure (3 dose) study in female albino rats with AD 32 administered by urinary bladder catheterization, 1991. SUBMITTED TO IND

B.2.2 Report WIL-178005. Repeated exposure (6 dose) study in female albino rats with AD 32 administered by urinary bladder catheterization, 1991. SUBMITTED TO IND

B.2.3 Report 3-E64. A repeated dose toxicity study of AD 32 administered inravesicularly to beagle dogs, 1997. SUBMITTED TO IND

B.2.4 Report OLI-RA247.06-9710-MJV-1, Thunberg AL. Summary report on the analysis of dosing solutions from toxicology study 3-E64 A repeated dose toxicity study of AD 32 administered intravesicularly to beagle dogs, 1997. Vol. 1.19, p 462

B.2.5 Report WIL-178003. Repeated exposure intraperitoneal toxicity study in female albino rats with AD 32,1991. SUBMITTED TO IND

B.2.6 Report 048-004. Repeated dose toxicity study of AD-32 administered intraperitoneally to Sprague-Dawley rats, 1997. SUBMITTED TO IND

B.2.7 Report OLI-RA247.06-9710-MJV-2, Thunberg AL. Summary report on the analysis of dosing solutions from toxicology study 048-004 Repeated dose toxicity study of AD 32 administered intraperitoneally to Sprague-Dawley rats, 1997. Vol. 1.20, p 484

B.2.8 Report 048-003. Repeated dose toxicity study of AD 32 administered intraperitoneally to beagle dogs, 1997. SUBMITTED TO IND

B.2.9 Report OLI-RA247.06-9710-NTJ-1, Thunberg AL. Summary report on the analysis of dosing solutions from toxicology study 048-003 Repeated dose toxicity study of AD 32 administered intraperitoneally to beagle dogs, 1997. Vol. 1.21, p 179

B.2.10 Report AD32RATCHSTX4. A multiple dose toxicity study of AD 32 injected directly into the thoracic cavity of rats, 1997. - SUBMITTED TO IND

B.2.11 Report AD32CHSTX4. A multiple dose(X4) toxicity study of AD 32 injected directly into the thoracic cavity of beagle dogs, 1996. SUBMITTED TO IND

B.3 Special Toxicity Studies

B.3.1 Report 048-001. Eye irritation study of AD 32 in New Zealand white (NZW) rabbits, 1996. SUBMITTED TO IND

B.3.2 Report 048-002. Primary dermal irritation study of AD 32 in New Zealand white (NZW) rabbits, 1996. SUBMITTED TO IND

B.4 Mutagenicity Studies

B.4.1 Report 3-D22.Evaluation of AD 32 in the Ames Assay With E. Coli, 1996. SUBMITTED TO IND

B.4.2 Report 3-D23. Evaluation of AD 32 in the in vitro chromosomal aberration assay, 1997. SUBMITTED TO IND

B.5 Non-GLP Supplemental Toxicity Studies

B.5.1 ReportNSC246131. Adriamycin cardiotoxicity. A comparison with adriamycin, AD-32 N-trifluoroacetyladriamycin-14-valerate, 1979. SUBMITTED TO IND

B.5.2 Report. Israel M, Sweatman TW and Parker RM. Histopathologic evaluation of rat bladder urothelium at 28 days post-intravesical instillation of NCI Diluent 12 vehicle and AD 32 formulated in NCI Diluent 12 vehicle, 1990. SUBMITTED TO IND

Appendix C Pharmacokinetic Study Reports

C.1 Report NIH-NO1-CM23242. Garattini S. Relationship of antineoplastic drug kinetics to in in vivo effects, 1997. SUBMITTED TO IND

C.2 Report. Israel M and Khetarpal VK. Comparative tissue distribution and metabolism studies with adriamycin and N-trifluoroacetyladriamycin-14-valerate (AD 32) in normal and tumor-bearing mice, 1991. SUBMITTED TO IND

C.3 Report. Sweatman TW and Israel M. Pharmacology of intraperitoneal AD 32 in the rat, 1991. SUBMITTED TO IND

C.4 Report. Sweatman TW. N-trifluoroacetyladriamycin-14-valerate (AD 32): drug penetration of the dog urinary bladder following intravesical administration, 1992. Vol. 1.24, p 380

C.5 Report. Sweatman TW. and Israel M. Validation report of the liquid chromatography assay for N-trifluoro-acetyladriamycin-14-valerate (AD 32) and principal biotransformation products in human serum, 1997. Vol. 1.24, p 390

C.6 Report. OLI-VRA595.1. Validation of a method for analysis of AD 32, AD 41 and AD 92 in human serum using following protein precipitation, 1997. Vol. 1.25, p 001

C.7 Report OLI-VRA595.2. Cross-validation of a method for analysis of AD 32, AD 41 and AD 92 in human serum to dog plasma, 1997. Vol. 1.25, p 092

C.8 Report OLI-RA595-9708-RAW-1. Analysis of AD 32, AD 41 and AD 92 in beagle dog plasma samples from Study 048-003, 1997. Vol. 1.25, p 238

C.9 Report OLI-RA595-9708-RAW-2. Analysis of AD 32, AD 41 and AD 92 in beagle dog plasma samples from Study 3-E64, 1997. Vol. 1.25, p 270

Appendix D Reference for Non-clinical

Studies 1.26001

Appendix E Individual AD 32 Published References, Vol. 1.26, p 017

1. Abbruzzi R. Rizzardini M, Benigni A, Barbieri B. Donelli, MG and Salmona M. Possible relevance of N-trifluoro-acetyladriamycin (AD41) in the antitumoral activity of N-trifluoroacetyladriamycin-14-valerate (AD 32) in tumor-bearing mice. I. Pharmacokinetic evidence. Cancer Treat Rep 1980; 64:873-8. Vol. 1.26, p 018

2. Ajito K, Atsumi S. Ikeda D, Kondo S. Takeuchi T and Umezawa K. Inhibition of human immunodeficiency virus-associated reverse transcriptase by 14-aacyladriamycins. J Antibiot 1989; 42:611-9. Vol. 1.26, p 024

3. Barbieri B,Abbruzzi R. BenigniA, Rizzardini M, Donelli MG, Garattini S et al. Quantitative thin layer chromatographic measurement of Ntrifluoroacetyl-adriamycin-14-valerate (AD 32) and trifluoroacetyl-adriamycin (AD 41) in blood and tissues. J Chromatography 1979; 163:195-200. Vol. 1.26, p 033

3a. Bekers O. Beiinen JH, Storm G. Bult A and Underberg WJM. Chemical stability of N-trifluoroacetyidoxorubicin-14-valerate (AD-32) in aqueous media and after liposome encapsulation. Int J Pharmaceut 1989; 56:103. Vol. 1.26, p 039

4. Blum RH, Garnick, MB, Israel M, Canellos GP, Henderson IC and Frei E lilt Preclinical rationale and Phase I clinical trial of the adriamycin analog, AD 32. In: Carter SK, Sakurai Y and Umezawa H. editors. New Drugs in Cancer Chemotherapy (Recent Results in Cancer Research, Volume 76). Berlin, Heidelberg, New York: Springer-Verlag, 1981: 7-15. Vol. 1.26, p 046.

5. Bodley A, Liu LF, Israel M, Seshadri R. Koseki Y. Giuliani FC et al. DNA topoisomerase 11 mediated interaction of doxorubicin and daunorubicin congeners with DNA. Cancer Res 1989; 49:5969-78. Vol. 1.26, p 055

6. Brox L, Gowans B. and Belch A. N-trifluoroacetyl-adriamycin-14-valerate and adriamycin-induced DNA damage in the RPMI 6410 human lymphoblastoid cell line. Can J Biochem 1980; 58:720-5. Vol. 1.26, p 065

7. Brox L, Gowans B. To R and Belch A. The effect of anoxia on anthracycline-induced DNA damage in the RPMI-6410 human lymphoblastoid cell line. Can J Biochem 1982; 60:873-6. Vol. 1.26, p 071

8. Burke TG, Doroshow JH and Tritton TR. Membrane interactions of the anthracycline antibiotics. SPIE-Int Soc Opt Eng 1988; 909:447-54. Vol. 1.26, p 075.

9. Burke TG, Israel M, Seshadri R. and Doroshow JH. A fluorescence study examining how 14 valerate side chain substitution modulates anthracycline binding to small unilamellar phospholipid vesicles. Biochim Biophys Acta 1989; 982:123-30. Vol. 1.26, p 084

10. Burke TG, Israel M, Seshadri R. and Doroshow JH. Abrogation of the selectivity of adriamycin for negatively-charged phospholipids by 14-valerate side chain substitution. Cancer Biochem Biophys 1990; 11:177-85. Vol. 1.26, p 092

11. Chow K-C, MacDonald TL and Ross WE. DNA binding by epipodophyllotoxins and N-acyl anthracyclines: Implications for mechanism of topoisomerase 11 inhibition. Molec Pharmacol 1988; 34:467-73. Vol. 1.26, p 101

12. Chuang LF, Kawahata RT and Chuang RY. - Inhibition of chicken myeloblastosis RNA polymerase 11 activity in vitro by N-trifluoroacetyladriamycin-14-valerate. FEBS Lett 1980; 117:247-51. Vol. 1.26, p 108

13. Chuang LF, Kung H-F, Israel M and Chuang RY. Activation of human leukemia protein kinase C by tumor promoters: Inhibition by N-trifluoroacetyladriamycin-14-valerate (AD-32). Biochem Pharmacol 1992; 43:865-72. Vol. 1.26, p 113.

14. Chuang RY, Chuang LF, Kawahata RT and Israel M. Effect of N-trifluoroacetyl-adriamycin 14-valerate on [^{3}H]thymidine uptake and DNA synthesis of human lymphoma cells. J Biol Chem 1983; 258:1062-5. Vol. 1.26, p 121

15. Constantinides PP, Inouchi N. Tritton T. Sartorelli AC, and Sturtevant JM. A scanning calorimetric study of the interaction of anthracyclines with neutral and acidic phospholipids alone and in binary mixtures. J Biol Chem 1986; 261: 10196-203. Vol. 1.26, p 125

16. Constantinides PP, Ghosaini L, Inouchi N. Kitamura S. Seshadri R. Israel M et al. Interaction of N-alkylanthracyclines with lipid bilayers: correlations between partition coefficients, lipid phase distributions and thermotropic behavior. Chem Phys Lipids 1989; 51:105-18. Vol. 1.26, p 133

17. Constantinides PP, Inouchi N. Sartorelli AC and Sturtevant JM. Interaction of adriamycin and N-trifluoroacetyladriamycin-14-valerate with cardiolipin-containing lipid bilayers. J Liposome Res 1989; 1:245-60. Vol. 1.26, p 147

18. Croce AC, Prosperi E, Supino R. and Bottiroli G. Anthracycline-induced inhibition of membrane permeability functions dependent on metabolic energy. Br J Cancer 1986; 54:943-50. Vol. 1.26, p 163

19. Dantchev D, Slioussartchouk V, Paintrand M, Bourut C, Hayat M and Mathe G. Ultra-structural study of cardiotoxicity and skin alterations in the golden hamster after treatment with 8 different anthracyclines. C R Soc Biol 1979; 173:394-413. Vol. 1.26, p 171

20. Dantchev D, Slioussartchouk V, Paintrand M, Bourut C, Mathe G and Macieira-Coelho A. Comparative cardiac and skin toxicity of 8 anthracyclines: adriamycin (ADM), daunorubicin (DRB), detorubicin (DTR), 4-epi-adriamycin (eADM), adriamycin hydrochloride (ADMh), rubidazone (RBZ), aclacinomycin (ACM) and AD 32. Proc Amer Assoc Cancer Res and ASCO 1979; 20:48 (abstract). Vol. 1.26, p 191

21. Dantchev D, Slioussartchouk V, Paintrand M, Hayat M, Bourut C and Mathe G. Electron microscopic studies of the heart and light microscopic studies of the skin after treatment of golden hamsters with adriamycin, detorubicin, AD-32 and aclacinomycin. Cancer Treat Rep 1979; 63:875-88. Vol. 1.26, p 192

22. Dantchev D, Slioussartchouk V, Paintrand M, Bourut C, Hayat M and Mathe G. Ultra- structural study of the cardiotoxicity and light microscopic findings of the skin after treatment of golden hamsters with seven different anthracyclines. Cancer Res 1980; 74:223-49. Vol. 1.26, p 206

23. Dantchev D, Paintrand M, Bourut C, Pignot I, Maral R and Mathe G. Comparative experimental study and evaluation of the degree of cardiotoxicity and alopecia of twelve different anthracyclines using the golden hamster model. In: Mathe G. Maral R and DeJager R. editors.

Anthracyclines: Current Status and Future Developments. New York: Masson, 1981: 25-36. Vol. 1.26, p 233

24. Dantchev D, Balercia G. Bourut C, Anjo A, Maral R. and Mathe G. Comparative microscopic study of cardiotoxicity and skin toxicity of anthracycline analogs. Biomed Pharmacother 1984; 38:322-8. Vol. 1.26, p 246

25. David J-C, BassezT, BonhommetM, and Rusquet R. Inhibition of DNA ligase from human thymocytes and normal or leukemic lymphoby antileukemic drugs. Cancer Res 1985; 45:2177-83. Vol. 1.26, p 253

26. DiDomenico D, Brown J. Shaw M, Rubenstein M and Guinan P. Intralesional AD 32 is efficacious in a prostate cancer model. Proc Amer Assoc Cancer Res 1996; 3:303 (abstract). Vol. 1.26, p 260

27. Eckle E, Kiefer E, Lauser G and Stezowski JJ. Molecular level studies with anthracyclines. Drugs Exptl Clin Res 1985; 11 :207-17. Vol. 1.26, p 261

28. FacchineKi T. Mantovani A, Cantoni R. Cantoni L, PantaroKo C, and Salmona M. Effect of daunomycin, adriamycin and its congener AD 32 on the activity of DNase I from bovine pancreas. Biochem Pharmacol 1977; 26:1953-4. Vol. 1.26, p 272

29. FacchineKiT, MantovaniA, Cantoni L, Cantoni R and Salmona M. Intercalation with DNA is a prerequisite for daunomycin, adriamycin and its congeners in inhibiting DNAase I. Chem-Biol Interactions 1978; 20:97-102. Vol. 1.26, p 274

30. FacchineKi T. Raz A, and Goldman R. A differential interaction of daunomycin, adriamycin, and N-trifluoro-acetyladriamycin-14 valerate with mouse peritoneal macrophages. Cancer Res 1978; 38:3944-9. Vol. 1.26, p 280

31. Fink Sl, Leo A, Yamakawa M, Hansch C and Quinn FR. The quantitative structure-selectivity relationship of anthracycline antitumor activity and cardiac toxicity. Farmaco-Ed Sci 1980; 35:965-79. Vol. 1.26, p 286

32. Fisher J. Ramakrishnan K and McLane KE. Complexation of anthracycline antibiotics by the apo egg white riboflavin binding protein. Biochemistry 1982; 21:6172-80. Vol. 1.26, p 301

33. Frei E lil, Schlossman S and Israel M. New approaches to the treatment of non-Hodgkin's lymphoma. Cancer Treat Rep 1977; 61 :1209-17. Vol. 1.26, p 310

34. Friedman HS, Bigner SH, Schold SC Jr, and Bigner DO. The use of experimental models of human medulloblastoma in the design of rational therapy. In: Walker MD and Thomas DOT, editors. Biol Brain Tumour, Proceedings of the Second International Symposium; 1986: 405 9.? Vol. 1.26, p 319

35. Friedman HS, Schold SC Jr, and Bigner DO. Chemotherapy of subcutaneous and intracranial human medulloblastoma xenografts in athymic nude mice. Cancer Res 1986; 46:224-8. Vol. 1.26, p 324

36. Gamba-Vitalo C, Blair OC, Tritton TR, Lane PA, Carbone R. and Sartorelli AC. Cytotoxicity and differentiating actions of adriamycin in WEHI-3B D+ leukemia cells. Leukemia 1987; 1:188-97. Vol. 1.26, p 329

37. Gambetta RA, Banfi P, Lanzi C, Franzi A, and Zunino F. Protein kinase C activation and lipid peroxidation by doxorubicin analogues. Tumori 1989; 75:358-61. Vol. 1.26, p 339

38. Ganapathi R. Grabowski D, Rouse W and Riegler F. Differential effect of the calmodulin inhibitor trifluoperazine on cellular accumulation, retention, and cytotoxicity of anthracyclines in doxorubicin (adriamycin)-resistant P388 mouse leukemia cells. Cancer Res 1984; 44:5056-61. Vol. 1.26, p 343.

39. Ganapathi R. Grabowski D, Schmidt H. Yen A and lliakis G. Modulation of adriamycin and N-trifluoroacetyl-adriamycin-14-valerate induced effects on cell cycle traverse and cytotoxicity in

P388 mouse leukemia cells by caffeine and the calmodulin inhibitor trifluoperazine. Cancer Res 1986; 46:5553-7. Vol 1.26, p 349.

40. Giavazi R. Scholar E, and Hart IR. Isolation and preliminary characterization of an adriamycin-resistant murine fibrosarcoma cell line. Cancer Res 1983; 43:2216-22. Vol. 1.26, p 354.

41. GoldinA, VendiKiJM, MacDonald JS, Muggia FM, Henney JE and Devita VT Jr. Current results of the screening program at the Division of Cancer Treatment, National Cancer Institute. EurJ Cancer 1981; 17:129-42. Vol. 1.26, p 361.

42. Goldman R. FacchineKi T. Bach D, Raz A, and Shinitzky M. A differential interaction of daunomycin, adriamycin and their derivatives with human erythrocytes and phospholipid bilayers. Biochim Biophys Acta 1978; 512:254-69. Vol 1.26, p 375.

43. Henderson IC, Billingham M, Israel M, Krishan A and Frei E lilt Comparative cardiotoxicity studies with adriamycin (ADR) and AD-32 in rabbits. Proc Am Assoc Cancer Ftes 1978; 19:158 (abstract). Vol. 1.26, p 391.

44. Iliakis G and Lazar W. Effect of sodium chloride concentration on adriamycin and N trifluoroacetyl-adriamycin-14-valerate (AD 32) induced cell killing and DNA damage in Chinese hamster V79 cells. Cancer Res 1987; 47:1853-8. Vol. 1.26, p 406.

45. Israel M, Modest, EJ and Frei E lilt, N-trifluoroacetyl-adriamycin-14-valerate, an analog with greater experimental antitumor activity and less toxicity than adriamycin. Cancer Res 1975; 35:1365-8. Vol. 1.26, p 412.

46. Israel M, Pegg WJ, Hirst M, Modest EJ and Frei E lilt Significant antitumor activity of N-trifluoroacetyl-adriamycin-14-valerate (AD 32) a new adriamycin analog. Proc Amer Assoc Cancer Res 1975; 16:116 (abstract). Vol. 1.26, p 416.

47. Israel M, Pegg WJ, Seshadri R. and Parker LM. N-trifluoroacetyladriamycin-14-valerate (AD 32): some in viva structure activity relationships. Fifth International Symposium on Medicinal Chemistry, Paris, France, July 1976 (abstract). Vol. 1.26, p 417.

48. Israel M. N-trifluoroacetyladriamycin-14-valerate (AD 32): development of a new potent antitumor agent. Alpha Academy of Pharmaceutical Sciences, 124th American Pharmaceutical Association Meeting, New York, NY, May 1977 (abstract). Vol. 1.26, p 418.

49. Israel M, Karkowsky AM and Pegg WJ. Use of liquid chromatography in tissue distribution studies with AD 32, a promising new anticancer agent. International Liquid Chromatography Symposium II, Boston, MA, Oct 1978 (abstract). Vol. 1.26, p 419.

50. Israel M, Pegg WJ end Wilkinson PM. Urinary anthracycline metabolites from mice treated with adriamycin and N-trifluoroacetyladriamycin-14valerate. J. Pharmacol Exp Ther 1978; 204:696-701. Vol. 1.26, p 420.

51. Israel M, Wilkinson PM and Osteen RT. Comparative pharmacology of adriamycin (ADR) and N-trifluoro-acetyladriamycin-14-valerate (AD 32) in the monkey. Eleventh Intentional Cancer Congress, Buenos Aires, Argentina, Oct. 1978 (abstract). Vol. 1.26, p 426.

52. Israel M, Wilkinson PM, Pegg WJ and Frei E lilt Hepatobiliary metabolism and excretion of adriamycin and N-trifluoroacetyladriamycin-14 valerate in the rat. Cancer Res 1978; 38:365-70. Vol. 1.26, p 428.

53. Israel M, Karkowsky AM and Khetarpal VK. Anthracycline fluorescence and radioactivity in tissues of mice one hour after treatment with [^{14}C]-labeled AD 32. Proc Amer Assoc Cancer Res and Amer Soc Clin Oncol 1979; 20:225 (abstract). Vol. 1.26, p 434.

54. Israel M, Khetarpal VK, Pegg WJ, Seshadri R and Potti PGG. Further studies on the pharmacology of AD 32, a novel adriamycin analog. International Liquid Chromatography Symposium lil, Boston, MA, Oct 1979 (abstract). Vol. 1.26, p 435.

55. Israel M, Pegg WJ, Wilkinson PM and Garnick MB. HPLC applications in the analysis of adriamycin and analogs in biological fluids. In: Hawk GL, editor. Biological/Biomedical Applications of Liquid Chromatography. New York and Basel: Marcel Dekker, 1979:413-28. Vol. 1.26, p 436.

56. Israel M, Karkowsky AM and Pegg WJ. Pharmacologic studies with radiolabeled N-trifluoroacetyladriamycin-14-valerate (AD 32): Comparison of total anthracycline fluorescence and radioactivity in mouse serum and urine. Cancer Chemother Pharmacol 1980; 4:79-82. Vol. 1.26, p 452.

57. Israel M, Khetarpal VK, Potti PGG and Seshadri R. Distribution and metabolism of N-trifluoroacetyl-adriamycin-14-valerate (AD 32) in tissues of A/JAX mice. Proc Amer Assoc Cancer Res and Amer Soc Clin Oncol 1980; 21:256 (abstract). Vol. 1.26, p 456.

58. Israel M, Wilkinson PM and Osteen RT. Pharmacology studies with adriamycin and N-trifluoroacetyladriamycin-14-valerate (AD 32) in cynomolgus monkeys: additional evidence for the absence of an adriamycin-prodrug mechanism for AD 32. In: Crooke ST and Reich SD, editors. Anthracyclines: Current Status and New Developments. New York: Academic Press, 1980: 431-44. Vol. 1.26, p 457.

59. Israel M, Karkowsky AM and Khetarpal VK.Distribution of radioactivity and anthracycline---fluorescence in tissues of mice one hour after [^{14}C]-labeled AD 32 administration. Cancer Chemother Pharmacol 1981; 6:25-30. Vol. 1.26, page 471.

60. Israel M and Idriss JM. Differential mechanism of DNA synthesis inhibition in vitro for N-trifluoroacetyladriamycin -14-valerate (AD 32) and adriamycin (ADR). Proc Amer Assoc Cancer Res 1982; 23:172 (abstract). Vol. 1.27, p 1.

61. Israel M and Idriss JM. A possible mechanistic basis for the growth inhibitory properties of N-trifluoroacetyladriamycin-14-valerate (AD 32), a non-DNA binding adriamycin analog. Thirteenth International Cancer Congress, Seattle, WA, Sep 1982 (abstract). Vol. 1.27, p 2.

62. Israel M and KhetarpalVK. Tissue distribution and metabolism of N-trifluoroacetyl-adriamycin 14-valerate (AD-32) compared with adriamycin in mice bearing established Lewis lung carcinomas. Thirteenth International Congress of Chemotherapy, Vienna, Austria, Aug-Sep1983 (abstract). Vol. 1.27, p 3.

63. Israel M, Seshadri R. SweatmanTW, Kirschenbaum S. Silber R. and Potmesil M. Structure and biological activity of N-substituted anthracyclines: correlation with DNA interaction. First Conference on DNA Topoisomerases in Cancer Chemotherapy, New York, NY, Nov 1986 (abstract). Vol. 1.27, p 7.

64. Israel M, Idriss JM, Koseki Y and Khetarpal VK. Comparative effects of adriamycin and DNA non-binding analogues on DNA, RNA and protein synthesis in vitro. Cancer Chemother Pharmacol 1987; 20:277-84. Vol. 1.27, p 15.

65. Israel M, Seshadri R. Koseki, Y. Sweatman TW and Idriss JM. Amelioration of adriamycin toxicity through modification of drug-DNA binding properties. Cancer Treat Rev 1987; 14:163-7. Vol. 1.27, p 23.

66. Israel M, Chengelis CP, Naas DJ and Sweatman TW. Absence of N-trifluoroacetyl~adriamycin-14 valerate (AD-32) toxicity following intraperitoneal (ip) injection in rats. Proc Amer Assoc Cancer Res 1993; 34:427 (abstract). Vol. 1.27, p 28.

67. Israel M, Bernacki RJ, and Kanter PM. Pharmacologic rationale for intraprostatic AD 32 (N-trifluoroacetyladriamycin-14-valerate). Proc Amer Assoc Cancer Res 1996; 37:372 (abstract). Vol. 1.27, p 48.

68. Johnson RK, Chitnis MP, Embrey WM and Gregory EB. In vivo characteristics of resistance and cross-resistance of an adriamycin-resistant subline of P388 leukemia. Cancer Treat Rep 1978; 62:1535-47. Vol. 1.27, p 49.

69. Johnson RK and Howard WS. Development and cross-resistance characteristics of a subline of P388 leukemia resistant to 4'-(9 acridinylamino)-methanesulfon-m-anisidide. Eur J Cancer Clin Oncol 1982; 18:479-87. Vol. 1.27, p 62.

70. Kacinski BM and Rupp WD. Interactions of the UVRABC endonuclease in viva and in vitro with DNA damage produced by antineoplastic anthracyclines. Cancer Res 1984; 44:3489-92. Vol. 1.27, p 71

71. Kanter PM, and Schwartz HS. Effects of N-trifluoroacetyladriamycin-14-valerate and related agents on DNA strand damage and thymidine incorporation in CCRF-CEM cells. Cancer Res 1979; 39:448-51. Vol 1.27, p 75.

72. Kanter PM and Schwartz HS. Quantitative models for growth inhibition of human leukemia cells by antitumor anthracycline derivatives. Cancer Res 1979; 39:3661-72. Vol. 1.27, p 79.

73. Karkowsky AM and Israel M. Serum levels and tissue distribution studies in mice with radiolabeled AD 32. Proc Amer Assoc Cancer Res and Amer Soc Clin Oncol 19,78; 19:157 (abstract). Vol. 1.27, p 91.

74. Kashfi K, Israel M, Sweatman TW, Seshadri R.and Cook GA. Inhibition of mitochondrial carnitine palmitoyltransferases by adriamycin and adriamycin analogues. Biochem Pharmacol 1990; 40:144148. Vol. 1.27, p 92.

75. KhetarpalVKandisraelM. Tissue distribution and metabolism of N-trifluoroacetyl-adriamycin 14-valerate (AD 32) in AJAX mice. Pharmacologist 1980; 22:291 (abstract). Vol. 1.27, p 100.

76. Khetarpal VK and Israel M. Metabolism and disposition of N-trifluoroacetyl-adriamycin-14 valerate (AD 32) in mice bearing established Lewis Lung carcinomas. Fed Proc 1982; 41:1572 (abstract). Vol. 1.27, p 101.

77 .Kohler G. Shen V and Peck WA. Adriamycin inhibits PTH-mediated but not PGE2-mediated stimulation of cyclic AMP formation in isolated bone cells. Calcif Tissue Int 1984; 36:279-84. Vol. 1.27, p 114.

78. Koseki Y. Sweatman TW, and Israel M. N-trifluoroacetyladriamycin-14-valerate (AD32) cellular pharmacology in human bladder tumor cell lines. ProcAmerAssoc Cancer Res 1992; 33:428 (abstract). Vol. 1.27, p 120.

79. Koseki Y. Sweatman TW, Savranskaya L, and Israel M. Comparative cytotoxicity of N trifluoroacetyladriamycin-14-valerate (AD-32), paclitaxel (TXL) and doxorubicin (DOX) vs. cultured human ovarian carcinoma cell lines: implications for AD 32 intraperitoneal (IP) therapy. Proc Amer Assoc Cancer Res 1995; 36:247 (abstract). Vol. 1.27, p 133.

80. Koseki Y. Savranskaya L, Samant S. Robbins KT, Los G and Sweatman TW. Comparative activity of N-trifluoroacetyladriamycin-14-valerate (AD 32) vs cisplatin in wild-type and platinum resistant human squamous carcinoma cell lines. Fourth International Conference on Head and Neck Cancer, Toronto, Canada, 1996 (abstract). Vol. 1.27, p 144,

80a. Koseki Y. Savranskaya L, Los G and Sweatman TW. In vitro synergism of N-trifluoroacetyladriamycin-14-valerate (AD 32) with low dose irradiation against perental and platinum-resistant squamous carcinoma cells. ProcAmerAssoc Cancer Res 1997; 38:331, (abstract). Vol. 1.27, p 159.

81. Krishan A, Israel M, Modest EJ and Frei E. Differences in cellular uptake and cytofluorescence of adriamycin and N-trifluoroacetyladriamycin-14-valerate. Cancer Res 1976; 36:2114-2116. Vol. 1.27, p 160.

82. Krishan A, Ganapathi RN and Israel M. Effect of adriamycin and analogs on the nuclear fluorescence of propidium iodide-stained cells. Cancer Res 1978; 38:3656-62. Vol. 1.27, p 163.

83. Krishan A and Ganapathi R. Laser flow cytometry and cancer chemotherapy: detection of intracellular anthracyclines by flow cytometry. J Histochem Cytochem 1979; 27:1655-6. Vol. 1.27, p 170.

84. Krishan A and Israel M. Excitation and quantitation of adriamycin and related anthracyclines by laser flow cytometry. Proc Amer Assoc Cancer Res and ASCO 1979; 20:220 (abstract). Vol. 1.27, p 172.

85. Krishan A and Ganapathi R. Laser flow cytometric studies on the intracellular fluorescence of anthracyclines. Cancer Res 1980; 40:3895-900. Vol. 1.27, p 173.

86. Krishan A and Ganapathi R. Laser flow cytomeric studies on the intracellular fluorescence of anthracyclines. Cancer Res 1908; 40:3895-900. Vol. 1.27, p 179.

87. Krishan A, Saueteig A, Gordon K and Swinkin C.Flow cytometric monitoring of cellular anthracycline accumulation in murine leukemic cells. Cancer Res 1986;46: 1768-73 Vol. 1.27, p 185.

88. Lampidis TJ, Johnson LV< Israel M and Canellos GP. Increased nuclear accumulation of adriamycin (ADR) and damage in cardiac muscle vs. Cardiac non-muscle cells in culture. Proc Amer Assoc Cancer Res and ASCO 1980; 21-275 (abstract). Vol. 1.27, p 191.

89. Lampidis TJ, Johnson LV and Israel M. Effects of adriamycin on rat heart cells in culture: increased accumulation and nucleoli fragmentation in cardiac muscle vs. Non-muscle cells. J Mol Cell Cardiol 1981; 13:913-24. Vol. 1.27, p 192.

90. Lazarus H. Yuan G. Tan E and Israel M. Comparative inhibitory effects of adriamycin, AD 32 and related compounds on in vitro cell growth and macromolecular syntheses. Proc Amer Assoc Cancer Res 1978; 19:159 (abstract). Vol 1.27, p 204.

91. Lehotay DC, Levey BA, Rogerson BJ and Levey GS. Inhibition of cardiac guanylate cyclase by doxorubicin and some of its analogs: possible relationship to cardiotoxicity. Cancer Treat Rep 1982; 66:311-6. Vol 1.27, p 205.

92. Levin M, Silber R. Israel M, Goldfeder A, Khetarpal VK and Potmesil M. Protein associated DNA breaks and DNA-protein cross links caused by DNA non-binding derivatives of adriamycin in L1210 cells. Cancer Res 1981; 41:1006-10. Vol. 1.27, p 211.

93. Lowe MC, Gram TE and Guarino AM. The utility of changes in cardiac weight as an index of drug-induced cardiotoxicity. J Environ Path Toxicol Oncol 1985; 6:57-68. Vol. 1.27, p 216.

94. Lown JW, Plambeck JA, Sondhi SM and Peters JH. Reactive oxygen species including superoxide ion generated from activated anthracyclines and the relationship to cardiotoxicity. In: RotilioG, editor. Superoxide and Superoxide Dismutase: In: Chemistry, Biology and Medicine, Proceedings from the Fourth International Conference; 1986:636-8. Vol. 1.27, p 227.

95. Maniar N. Krishan A, Israel M and Sam TSA. Anthracycline-induced DNA breaks and resealing in doxorubicin-resistant murin leukemic P388 cells. Biochem Pharmacol 1988; 37:1763-72. Vol. 1.27, p 230.

96. Mantovani A, Luini W. Peri G. Vecchi A and Spreafico F. Effect of chemotherapeutic agents on natural cell-mediated cytotoxicity in mice. Vol. 1.27, p 238.

97. Marsh J. Comparison of the sensitivities of human, canine and murine hematopoietic precursor cells to adriamycin and N-trifluoroacetyladriamycin-14-valerate. Cancer Res 1979; 39:360-4. Vol. 1.27, p 245.

98. Marsh JO. Correlation of hematologic toxicity of antineoplastic agents with their effects on bone marrow stem cells; interspecies studies using an in viva assay. Exp Hematol 1985; 13 (suppl 16):16-22. Vol. 1.27, p 250.

99. Marzin D, Jasmin Ć, Maral R and Mathe G. Mutagenicity of eight anthracycline derivatives in five strains of *Salmonella typhimurium*. Eur J Cancer Clin Oncol 1983; 19:641 -7. Vol. 1.27, p 257.

100. Masquelier M, Vitols S. Rudling M and Peterson C. Incorporation and binding of anthracycline derivatives to low density lipoprotein: in vitro and in viva studies on drug-LDL conjugates. In: Ishigami J. editor. Recent Advances in Chemotherapy, Proceedings of the 14th International Congress of Chemotherapy. 1985:311-2. Vol. 1.27, p 264.

101. Masquelier M, Vitols S. and Peterson C. Low-density lipoprotein as a carrier of antitumoral drugs: in viva fate of drug-human low-density lipoprotein complexes in mice. Cancer Res 1986; 46:3842-7. Vol. 1.27, p 266.

102. Muggia F. Levin M, Israel M, Potmesil M and Silber R. Comparative effects of AD-32 and adriamycin on DNA. Fifth Annual Meeting of the Medical Oncology Society, Nice, France, Dec 1979 (abstract). Vol. 1.27, p 272.

103. Nair S. Singh SV, Samy TSA, and Krishan A. Anthracycline resistance in murine leukemic P388 cells: role of drug efflux and glutathione related enzymes. Biochem Pharmacol 1990; 39:723-8. Vol. 1.27, p 273.

104. Niell HE, Hunter RF, Herrod HO and Israel M. The activity of adriamycin (ADR) and N-trifluoroacetyl-adriamycin-14-valerate (AD 32) against human bladder tumor cell lines (HBTCL). Proc Amer Assoc Cancer Res 1986; 27:412 (abstract). Vol. 1.27, p 279.

105. Niell HE, Hunter RF, Herrod HO and Israel M. Effects of N-trifluoro-acetyladriamycin-14 valerate (AD-32) on human bladder tumor cell lines. Cancer Chemother Pharmacol 1987; 19:47-52. Vol. 1.27, p 280.

106. Parker LM, Israel M, Hirst M, Modest EJ and Frei E lilt Further in viva studies with N-trifluoroacetyladriamycin-14-valerate (AD 32). Proc Amer Assoc Cancer Res 1976; 17:108 (abstract). Vol. 1.27, p 286.

107. Parker LM, Hirst M and Israel M. N-trifluoroacetyl-adriamycin-14-valerate: additional mouse antitumor and toxicity studies. Cancer Treat Rep 1978; 62:119-127. Vol. 1.27, p 287.

108. Pearlman LF, Chuang RY, Israel M and Simpkins H. Interaction of three second generation anthracyclines with polynucleotides, RNA, DNA, and nucleosomes. Cancer Res 1986; 46:341-6. Vol. 1.27, p 296.

109. Pegg WJ, Wilkinson PM and Israel M. Comparative anthracycline metabolite patterns in urine of mice treated with adriamycin (ADR) and N-trifluoroacetyladriamycin-14-valerate (AD 32). 173rd National Meeting, American Chemical Society, New Orleans, La., March 1977 (abstract). Vol. 1.27, p 302.

110. Peters JH, Gordon GR and Kashiwase D. Redox activities of antitumor anthracyclines determined by microsomal oxygen consumption and assays for superoxide anion and hydroxyl radical eneration. Biochem Pharmacol 1986; 35:1309-23. Vol. 1.27, p 303.

111. Potmesil M, Levin M, Israel M and Silber R. Protein-associated DNA breaks caused by adriamycin or N-trifluoroacetyladriamycin-14 valerate in vivo: relationship to cell clonogenicity. In: Periti P and Grassi GG, editors. Current Chemotherapy and Immunotherapy, Volume 2, American Association for Microbiology, Washington, DC. 1982: 1439-41. Vol. 1.27, p 318.

112. Potmesil M, Kirschenbaum S. Israel M and Silber R. Impaired repair of radiation-induced DNA damage in L1210 cells pretreated with DNA non-binding analogs of adriamycin. International Congress of Radiation Research, Amsterdam, The Netherlands, June 1983 (abstract). Vol. 1.27, p 321.

113. Potmesil M, Levin M, Traganos F. Israel M, Darzynkiewicz Z. Khetarpal VK and Silber R. In vivo effects of adriamycin or N-trifluoroacetyladriamycin-14-valerate on a mouse lymphoma. Eur J Cancer Clin Oncol 1983; 19:109-22. Vol. 1.27, p 323.

114. Potmesil M, Israel M, Seshadri R. Sweatman TW, Kirschenbaum S and Silber R. Structure and biological activity of N-substituted anthracyclines: correlation with drug-DNA topoisomerase 11 interaction. Proc Amer Assoc Cancer Res 1987; 28:262 (abstract). Vol. 1.27, p 337.

115. Prabhakar YS, Gupta SP and Ray A. Effect of molecular size on the activity of adriamycin analogs: a quantitative structure-activity relationship study. J Pharmacobio-Dyn 1986; 9:61-7. Vol. 1.27, p 346.

116. Pratesi G. Casazza AM, and Di Marco A. Antitumor activity of N-trifluoro-acetyladriamycin 14-valerate. Cancer Treat Rep 1978; 62:105-10. Vol. 1.27, p 353.

117. Raz A. B16 melanoma cell variants: irreversible inhibition of growth and induction of morphologic differentiation by anthracycline antibiotics. JNCI 1982; 68:629-33. Vol. 1.27, p 359.

118. Schold SC Jr, Friedman HS and Bigner DD. Therapeutic profile of the human glioma line D-54 MG in athymic mice. Cancer Treat Rep 1987; 71:849-50. Vol. 1.27, p 364.

119. Schwartz HS and Kanter PM. Biochemical parameters of growth inhibition of human leukemia cells by antitumor anthracycline agents. Cancer Treat Rep 1979; 63:821 -5. Vol. 1.27, p 366.

120. Schwartz HS and Kanter PM. Chemical interactions of cardiolipin with daunorubicin and other intercalating agents. Eur J Cancer 1979; 15:923-8. Vol. 1.27, p 371.

121. Schwartz HS and Kanter PM. DNA damage by anthracycline drugs in human leukemia cells. Cancer Lett 1981; 13:309-13. Vol. 1.28, p 1.

122. Seeber S. Loth H. and Crooke ST. Comparative nuclear and cellular incorporation of daunorubicin, doxorubicin, carminomycin, marcellomycin, aclacinomycin A, and AD 32 in daunorubicin-sensitive and -resistant Ehrlich Ascites in vitro. J Cancer Res Clin Oncol 1980; 98:109-18. Vol. 1.28, p 6.

123. Seeber S. Osieka R. Schmidt CG, Achterrath W and Crooke ST. In vivo resistance towards anthracyclines, etoposide and cis diaminodichloroplatinum (II). Cancer Res 1982; 42:4719-25. Vol. 1.28, p 16.

124. Sengupta SK, Seshadri R. Modest EJ and Israel M. Comparative DNA-binding studies with adriamycin (ADR), N-trifluoroacetyladriamycin 14-valerate (AD 32) and related compounds. Proc Amer Assoc Cancer Res 1976; 17:109 (abstract). Vol. 1.28, p 23.

125. Siegfried JM, Tritton TM, and Sartorelli AC. Comparison of anthracycline concentrations in S180 cell lines of varying sensitivity. Eur J Cancer Clin Oncol 1983; 19:113341. Vol. 1.28, p 24.

126. Silber R. Liu LF, Israel M, Bodley AL, Hsiang Y- H. Kirschenbaum S et al. Metabolic activation of N-acyl-anthracyclines precedes their interaction with DNA topoisomerase 11. NCI Monogr 1987; 4:111-5. Vol. 1.28, p 33.

127. Simpkins H. Figliomeni M and Rosen M. The interaction of platinum and anthrapyrazole antitumor drugs with mouse thymocytes studied by terbium fluorescence. Biochim Biophys Acta 1988; 972:25-32. Vol. 1.28, p 39.

128. Slovak ML, Hoeltge GA, and Ganapathi R. Abnormally banded chromosomal regions in doxorubicin-resistant B16-BL6 murine melanoma cells. Cancer Res 1986; 46:4171 -7. Vol. 1.28, p 47.

129. Smith RG. Characterization of anthracycline antibiotics by desorption chemical ionization mass spectrometry. Anal Chem 1982; 54:2006-8. Vol. 1.28, p 54.

130. Sterrenberg L, Julicher RHM, Goossens PAL, Bast A and Noordhoek J. Non-enzymatic lipid peroxidation in microsomes and microsomal phospholipids induced by anthracyclines. Free Rad Res Comms 1986; 1:369-78. Vol. 1.28, p 57.

131. Sun IL and Crane FL. Effects of anthracycline compounds on transmembrane redox function of cultured HeLa cells. Proc Indiana Acad Sci 1984; 93:267-74. Vol. 1.28, p 67.

132. Sun IL, Crane FL, Low H and Grebing C. Inhibition of plasma membrane NADH dehydrogenase by adriamycin and related anthracycline antibiotics. J Bioenerg Biomembr 1984; 16:209-21. Vol. 1.28, p 75.

133. Sweatman W and Israel M. Pharmacology of N-trifluoroacetyladriamycin-14-valerate (AD 32) administered to rats by intravesical instillation. Proc Amer Assoc Cancer Res 1989; 30:600 (abstract). Vol. 1.28, p 88.

134. Sweatman TW and Israel M. Effect of surfactant formulation (NCI Diluent 12) on intravesical drug pharmacology. J Cancer Res Clin Oncol 1990; 116 (suppl 1):538 (abstract). Vol. 1.28, p 103.

135. Sweatman W and Israel M. Pharmacology of intravesical (IVE) N-trifluoroacetyl-adriamycin 14-O-hemiadipate (AD143) in the rat. Proc Amer Assoc Cancer Res 1990; 31:382 (abstract). Vol. 1.28, p 105.

136. Sweatman TW, Parker RF and Israel M. Pharmacologic rationale for intravesical N trifluoroacetyladriamycin-14-valerate (AD-32): a preclinical study. Cancer Chemother Pharmacol 1991; 28:1-6. Vol. 1.28, p 121.

137. Sweatman W and Israel M. Pharmacology of intraperitoneal (IP) N-trifluoro-acetyladriamycin 14-valerate (AD-32) in the rat. Proc Amer Assoc Cancer Res 1992; 33:520 (abstract). Vol. 1.28, p 127.

138. Sweatman TW, Payne C, Eltorky MA, and Israel M. Drug penetration into dog bladder wall following intravesical (IVE) instillation of N-trifluoroacetyl-adriamycin-14-valerate (AD 32). Proc Amer Soc Cancer Res 1993; 34:364 (abstract). Vol. 1.28, p 149.

139. Sweatman TW and Israel M. Anthracyclines. In: Teicher BA, editor. Cancer Therapeutics: Experimental and Clinical Agents. Part 1: Cytotoxic Agents. Totowa, NJ: Humana, 1996: 113-136. Vol. 1.28, p 164.

140. Tomlinson E and Malspeis L. Concomitant adsorption and stability of some anthracycline antibiotics. J Pharm Sci 1982; 71:1121-5. 1.28, p 188.

141. Vecchi A, Cairo M, Mantovani A, Sironi M and Spreafico F. In vitro and in vivo studies on antitumoral activity of N-trifluoroacetyladriamycin-14-valerate (AD 32). Tenth International Congress of Chemotherapy, Zurich, Swikerland, Sep 1977 (abstract). Vol. 1.28, p 193.

142. Vecchi A, Cairo M, Mantovani A, Sironi M and Spreafico F. Comparative antineoplastic activity of adriamycin and N-trifluoroacetyladriamycin 14-valerate. Cancer Treat Rep 1978; 62:111 -7. Vol. 1.28, p 200.

143. Vecchi A, Spreafico F. Sironi M, Cairo M and Garattini S. The immunodepressive and hematotoxic activities of N-trifluoroacetyladriamycin-14-valerate. Eur J Cancer 1980; 16:1289-96. Vol. 1.28, p 207.

144. Vitols S. Gahrton G and Peterson C. Significance of the low-density lipoprotein (LDL) receptor pathway for the in vitro accumulation of AD-32 incorporated into LDL in normal and leukemic white blood cells. Cancer Treat Rep 1984; 68:515-20. Vol. 1.28, p 215.

145. Waldes H and Center M. Adriamycin-induced compaction of isolated chromatin. Biochem Pharmacol 1982; 31:1057-61. Vol. 1.28, p 221.

146. Wecker MT and Myers-Robfogel MW. DNA non-binding adriamycin analogs induce erythroid differentiation of K562 cells. Proc Amer Assoc Cancer Res 1984; 25:299 (abstract). Vol. 1.28, p 226.

147. Wilkinson PM, Israel M, Pegg WJ and Frei E lilt Comparative hepatobiliary metabolism and excretion of adriamycin (ADR) and N-trifluoroacetyladriamycin-14-valerate (AD 32). Proc Amer Assoc Cancer Res 1977; 18:188 (abstract). Vol. 1.28, p 227.

148. Wilkoff LJ, and Dulmadge EA. Resistance and cross-resistance of cultured leukemia P388 cells to vincristine, adriamycin, adriamycin analogs, and actinomycin D. JNCI 1978; 61:1521-24. Vol. 1.28, p 228.

149. Wilson RG, King M, Lockwood R and McNeil M. Effects of N-trifluoroacetyladriamycin-14-valerate (AD 32) on nuclear RNA synthesis in cultured L1210 cells. Chem Biol Interact 1980; 32:331-7. Vol. 1.28, p 232.

150. Yesair DW and Israel M. In vitro metabolism of some N-substituted adriamycin analogues. Proc Amer Assoc Cancer Res 1975; 16;115 (abstract). Vol. 1.28, p 239.

151. Young RC, Ozols RG and Myers CE. The anthracycline antineoplastic drugs. New England J Med 1981; 305:139-53. Vol. 1.28, p 240.

152. Zbinden G. Bachman M and Holderegger C. Model systems of cardiotoxic effects of anthracyclines. Antibiotics Chemother 1978; 23:255-70. Vol. 1.28, p 255.

153. Zini G. Vicario GP, Lazzati M, and Arcamone F. Radioactive species in rat urine and tissues after [^{14}C] AD 32 administration. Cancer Chemother Pharmacol 1984; 12:154-6. Vol. 1.28, p 271.

154. Zirvi KA, van der Bosch J and Kaplan NO. In vitro responses of nude mouse-xenografted human colon carcinomas exposed to doxorubicin derivatives in tissue culture and in the mouse. Cancer Res 1982; 42:3793-7. Vol. 1.28, p 274.

155. Zirvi KA, Masui H. Giuliani FC and Kaplan NO. Correlation of drug sensitivity on human colon adenocarcinoma cells grown in soft agar and in athymic mice. Int J Cancer 1983; 32:45-51. Vol. 1.28, p 279.

156. Zirvi KA, Gilani SH and Hill GJ. Embryotoxic effects of doxorubicin and N-trifluoroacetyl adriamycin-14-valerate(AD-32). Teratology 1985; 31:247-52. Vol. 1.28, p 286.

6.10.1.1 Published Studies and Abstracts, Intravesical Administration, Vol. 1.31, p 134

Israel M, Sweatman TW, Soloway MS, Makkin H. Rangel MOM, Eltorky MA. Initial clinical Pharmacology of intravesical (IVE) AD 32 [abstract]. Proc Amer Assoc Cancer Res 1992;33:445.

6.10.2 Intravenous Administration-

Israel M. Garnick MB, Pegg WJ, Blum RH, Frei E lilt Preliminary pharmacology of AD 32 in Man [abstract 637]. Proc Amer Assoc Can Res and ASCO 1978; 19:16.

6.10.2.2 Published Studies and Abstracts, intravenous Administration. Vol. 1.31, p 163.

Garnick MB, Israel M, Pegg WJ, Blum RH, Smith E, Frei E lilt Hepatobiliary Pharmacokinetics of AD 32 in an [abstract 834]. Proc Amer Assoc Cancer Res and ASCO 1979;20:206.

6.10.2.3 Published Studies and Abstracts, Intravenous Administration. Vol. 1.31, p 172.

Blum RH, Garnick MB, Israel M, Canellos GP, Henderson IC, Frei E lilt Preclinical rationale and Phase I clinical trial of the adriamycin analog, AD 32. In: Carter SK, Sakurai Y. Umezawa H. editors. New drugs in cancer chemotherapy (Recent results in cancer research, Volume 76). Berlin: Springer-Verlag 1981:7-15.

6.10.2.4 Published Studies and Abstracts, Intravenous Administration. Vol. 1.31, p 186

Garnick MB, Griffin JO, Sack MJ, Blum RH, Israel M, Frei E lilt Phase II evaluation of N trifluoroacetyladriamycin-14-valerate (AD-32). In: Muggia FM, Young CW, Carter SK, editors. Anthracycline antibiotics in cancer therapy (Developments in oncology, Volume 10). The- Hague: Martinus Hijhoff, 1982:541-8.

6.10.3 Pre-clinical Intravesical Administration

6.10.3.1 Published Studies and Abstracts, Pre-clinical Intravesical Administration, Vol. 1.31, p 201

Sweatman NEW, Parker RF, Israel M. Pharmacologic rationale for Intravesical N trifluoroacetyladriamycin-14 valerate (AD 32): a Pre-clinical study. Cancer ChemOther Pharmaco 1991 ;28: 1-6

6.10.4 Structurally Related Drugs: Doxorubicin Hydrochloride Intravesical Administration

6.10.4.1 Published Studies and Abstracts, Doxorubicin Hydrochloride Intravesical Administration

Chai M, Wientjes MG, Badalament RA, et al. Pharmacokinetics of Intravesical doxorubicin in superficial bladder cancer patients. J Urology 1994; 152:374-8. Vol. 1.31, p 214.