August 1, 1995
Ed Fisher, Ph D.

Review and Evaluation of Pharmacology and Toxicology

NDA: 20-505

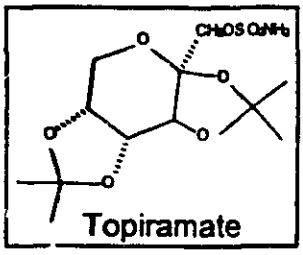
Sponsor: The RW Johnson Pharmaceutical Research Institute
Spring House, PA 19477

Drug: Topamax (topiramate)

Chemical Name: [2,3:4,5-bis-O-(1-methylethylidene)-β-D-fructopyranose sulfamate]

Code name(s): RWJ-17021-000, McN-4853, KW-6485

Structure:



Mol. formula: $C_{12}H_{21}NO_8S$

Mol. Wt.: 339.36

Category: Antiepileptic

Related IND(s):

Table of Contents

I. PHARMACODYNAMICS

A) ANTICONVULSANT SCREENING IN MOUSE AND RAT (McNeil studies, A44206, Vol. 17)

1. Topiramate was active against MES-induced seizures; the oral MES ED50 was 36.5 (23.7-56.2) mg/kg in mice and 17.5 (13.5-22.6) mg/kg in rats.
2. It was not active versus PTZ-, Pic-, Bic-, and Strych-induced seizures at 2X MES ED50.
3. The duration of anticonvulsant action after oral administration was relatively long (>8 hr).
4. The PI was 30 in mice at 4 hr after oral administration using MES and rotorod neurotoxicity.
5. No apparent loss of anticonvulsant activity was seen after po administration of topiramate (30 mg/kg po) or acetazolamide (30 mg/kg po) for 14 days in rats.
6. Topiramate was less potent than acetazolamide in inhibiting carbonic anhydrase (previous data), but had similar anticonvulsant potency.
7. Reserpine abolished the anti-MES activity of both topiramate and acetazolamide.
8. SKF-525A significantly increased the anticonvulsant potency of topiramate.

B) ANTICONVULSANT SCREENING IN MOUSE AND RAT studies, A41941, Vol. 17)

1. Topiramate was relatively potent against MES-induced seizures in mice and rats, but elevated the threshold for minimal Metrazol seizures only at about 2X the TD50 for minimal neurotoxicity (400 mg/kg).
2. It was inactive against sc Bic-, Pic-, and sc Strych-induced seizures at up to 500 mg/kg.
3. Decreased motor activity, rotorod toxicity, ataxia, and decreased respiration were seen at 1TD50. Sedation, ptosis, muscle relaxation, loss of righting reflex, analgesia, anesthesia, and death were seen at 4xTD50.
4. PI values, after ip administration in mice or oral administration in mice and rats, were at least 2X those for prototype AEDs. (PI for oral PHT in rats invalidated by dose-dependent absorption.)
5. Complete oral absorption was suggested by po/ip ratios for MES ED50 (0.56) and rotorod TD50 (0.97) in mice.
6. Conclusions: favorable absorption and safety ratios; overall profile similar to that of phenytoin.

C) COMPARISON OF ANTICONVULSANT ACTIVITY OF TOPIRAMATE WITH THAT OF REFERENCE AGENTS (A49951, Vol. 17)

1. The anticonvulsant profile of topiramate was similar to those of phenytoin and carbamazepine, ie, active against MES-induced seizures but not against chemoconvulsant-induced seizures.
2. In the mouse MES test, topiramate (po ED50 = 43.8 mg/kg) was less potent than phenytoin (11.6), carbamazepine (16.4), and phenobarbital (21.5), more potent than VPA (802.6), and equipotent with acetazolamide **(Table I.1)**.
3. In the rat MES test, topiramate (po ED50 = 17.3 mg/kg) was equipotent with carbamazepine, phenobarbital, and acetazolamide, and was more potent than phenytoin (32.9). **(Table I.2)**
4. Topiramate had a high toxicity/anticonvulsant activity ratio, comparing favorably with the other compounds tested **(Tables I.3 & I.4)**.

Table I.1: Anticonvulsant Activity of Topiramate and Standard AEDs in the Mouse

Test	Route	ED50 mg/kg (95% Confidence Limits)					
		Topiramate	Phenytoin	Phenobarbital	Carbama-zepine	VPA	Acetazol-amide
MES	po	43.8 (30.5-60.9)	11.6 (8.6-14.4)	21.5 (17.9-25.4)	16.4 (12.1-23.3)	802.6 (721.7-918.4)	35.4 (24.8-48.9)
	ip	38.6 (28.3-49.1)	8.4 (6.9-10.2)	16.4 (13.2-19.9)	18.1 (15.6-21.2)	419.4 (348.6-486.4)	42.8 (35.0-51.7)
Metrazol	po	1026.5 (872-1184)	>120	18.7 (14.0-24.5)	>240	482.1 (426.4-530.5)	40% block @ 1000
	ip	1127.1 (966-1330)	—	—	—	—	40% block @ 1000

Table I.2: Anticonvulsant Activity of Topiramate and Standard AEDs in the Rat

Test	Route	ED50 mg/kg (95% Confidence Limits)				
		Topiramate	Phenytoin	Phenobarbital	Carbama-zepine	Acetazol-amide
MES	po	17.3 (9.9-23.6)	32.9 (31.3-44.1)	15.7 (7.2-25.4)	25.9 (19.2-37.0)	18.6 (8.9-28.0)
	ip	24.5 (16.3-42.9)	27.6 (19.3-43.9)	—	—	—
Metrazol	po	>1000	>1000	34.6 (27.4-41.6)	>1000	—
	ip	>1000	—	—	—	—

Table I.3: Ratio of Toxicity ED50's to MES ED50's for Topiramate and Standard AEDs in the Mouse

	Route	Topiramate	Phenytoin	Pheno-barbital	Carbama-zepine	Acetazol-amide
LD50/ MES ED50	po	>41	13.8	10.0	63.5	18.6 (8.9-28.0)
	ip	>47	—	—	—	—
LRR ED50/ MES ED50	po	31.1	>21.5	8.1	43.6	
	ip	35.9	—	—	—	
MN ED50/ MES ED50	po	9.3	4.2	2.1	7.1	56
	ip	9.9	—	3.3	8.2	>47

LRR = Loss of righting reflex; MN = Minimal neurotoxicity in the Rotorod test

Table I.4: Ratio of Toxicity ED50's to MES ED50's for Topiramate and Standard AEDs in the Rat

	Route	Topiramate	Phenytoin	Pheno-barbital	Carbama-zepine	Acetazol-amide
LD50/ MES ED50	po	>116	>53	19	>77	>108
	ip	>61	17.6	—	—	—
LRR ED50/ MES ED50	po	>116	>53	12.5	>77	>108
	ip	>61	—	—	—	—
MN ED50/ MES ED50	po	>116	>53	3.5	20.8	>108
	ip	65	4.7	—	—	—

LRR = Loss of righting reflex; MN = Minimal neurotoxicity in the Rotorod test

D) PHARMACOLOGICAL PROFILE IN MOUSE, RAT, AND DOG
A500679, Vol. 17)

1. Topiramate at 100 mg/kg po or more completely inhibited electroshock seizures in mice but did not block PTZ seizures at doses up to 1000 mg/kg po.
2. Pentobarbital sleep time was dose-dependently prolonged at 3-300 mg/kg po in mice.
3. Doses of 1-10 mg/kg iv produced no significant effect on cerebral pH; acetazolamide decreased pH at 10 and 30 mg/kg iv.
4. A diuretic effect was seen at 100 mg/kg po or more in rats. Urine pH became alkaline at 30 mg/kg po or more. Acetazolamide produced slight diuresis at 10 mg/kg po.
5. Gastric pH was increased at 1000 mg/kg po in rats. Gastric secretion and total acid output were decreased at 300 mg/kg po or more. Blood gastrin was unaffected by doses up to 1000 mg/kg. Charcoal meal passage through small intestine decreased at 1000 mg/kg in mice. Mucosal bleeding in the stomach was produced at 300 and 1000 mg/kg po in rats.
6. No effects on spontaneous EEGs or spinal reflexes in rats at doses of 1-10 mg/kg iv.
7. No effects on BP or HR in unanesthetized rats at 30-1000 mg/kg po.
8. No effects on respiration, BP, HR, blood flow, or lead II EEG in anesthetized beagle dogs at 1, 3, and 10 mg/kg iv.
9. No effect on right atrial sinus rate or on contractile force of electrically driven left atria in guinea pig.
10. No effects on autonomic system, isolated smooth muscle, blood functions, hemolysis, or blood glucose.

E) EFFECTS ON MES SEIZURES AND BRAIN CARBONIC ANHYDRASE ACTIVITY IN DBA AND C57 MICE A500840, Vol. 18)

Topiramate and acetazolamide were examined for anticonvulsant activity (against MES) in seizure-prone DBA mice (abnormally high carbonic anhydrase activity in brain), and in C57 mice (not seizure-prone; normal brain CA activity).

1. MES ED50 (ip) values for both topiramate and acetazolamide were higher in the DBA strain; mean values for topiramate were 54.4 and 39.1 mg/kg in DBA and C57 mice, respectively; corresponding values for acetazolamide were 45 and 25.4 mg/kg.
2. In topiramate-treated mice (20 mg/kg ip), CA activity was inhibited by about 60% in whole blood, 90% in brain homogenates, and 80% in cytosol of whole brain tissue in both strains,

but only slightly inhibited in mitochondria (both strains), microsomes (DBA only), and synaptosomes (C57only). Acetazolamide was not tested for CA inhibition.

F) INHIBITION OF MES SEIZURES IN MICE: TOLERANCE AND CROSS-TOLERANCE STUDIES OF TOPIRAMATE AND ACETAZOLAMIDE (A500842, Vol. 18)

Mice were given vehicle or fixed doses of acetazolamide (200 mg/kg ip) or topiramate (160 mg/kg ip) once daily for 5 days (doses considered approximately 4X MES ED50s). After 48 or 72 hr washout periods, topiramate or acetazolamide was administered ip and tested for activity in the MES test. Test drug was administered 1 hr before MES testing.

1. Tolerance developed to anti-MES activity of acetazolamide; acetazolamide ED50 values were greater in acetazolamide-pretreated than in vehicle-pretreated mice (**Table I.5A**).
2. No cross-tolerance to topiramate was seen in acetazolamide-pretreated mice; topiramate ED50s were similar in vehicle- and acetazolamide-pretreated mice (**Table I.5A**).
3. After topiramate pretreatment, differences in topiramate ED50s between vehicle- and topiramate-pretreated mice indicated tolerance development (**Table I.5B**).
4. Cross-tolerance to acetazolamide was seen in topiramate-pretreated mice; acetazolamide ED50's were higher in topiramate-pretreated mice (**Table I.5B**).

Table I.5A: MES ED50's in Mice Pretreated with Acetazolamide (200 mg/kg ip) or Vehicle for Five Days

	ED50 (95% fiducial limits) mg/kg	
Pretreatment/Test Drug	48 hr	72 hr
Vehicle/Topiramate	NT	70.7 (52.9-98.2)
Acetazolamide/Topiramate	NT	81.1 (61.2-98.7)
Vehicle/Acetazolamide	89.1 (66.8-128)	88.0 (67.3-108)
Acetazolamide/Acetazolamide	134 (108-219)*	382 (227->1000)*

* $p \leq 0.05$

Table I.5B: MES ED50's in Mice Pretreated with Topiramate (160 mg/kg ip) or Vehicle for Five Days

	ED50 (95% fiducial limits) mg/kg	
Pretreatment/Test Drug	48 hr	72 hr
Vehicle/Topiramate	55.1 (41.8-66.4)	54.5 (44.5-65.2)
Topiramate/Topiramate	80.8 (68.1-103)*	71.2 (51.5-92.3)
Vehicle/Acetazolamide	75.7 (52.9-94.2)	77.8 (59.0-99.2)
Acetazolamide/Acetazolamide	98.9 (81.6-125)	382 (101-318)*

* $p \leq 0.05$

G) ANTICONVULSANT ACTIONS IN SPONTANEOUSLY EPILEPTIC RATS AND DBA/2 MICE A505012, A505013, Vol. 18)

1. Topiramate (10, 20, 40 mg/kg ip) inhibited both tonic and absence-like seizures in SERs in a dose-dependent manner; phenytoin (15, 20 mg/kg), and zonisamide (20, 40 mg/kg) inhibited only the tonic seizure component.
2. Inhibitory effect of topiramate on absence-like seizures in SERs was antagonized by haloperidol (0.5 mg/kg ip), but activity against tonic seizures was unaffected.
3. Basal levels of glutamate and aspartate in dialysates of SER hippocampus were about 2X higher than in normal Wistar rats. Topiramate (20 mg/kg ip) reduced levels of both EAAs (45% below baseline) with a time course parallel to that for suppression of tonic seizures. EAA levels in Wistar were not affected by topiramate.
3. Topiramate inhibited sound-induced seizures in DBA/2 mice (ED50 = 8.6 mg/kg po).

H) BROAD PHARMACOLOGICAL EVALUATION OF TOPIRAMATE (A44488, Vol. 18)

1. MTD (po) in dogs was between 100 and 500 mg/kg; convulsions were seen in 1/3 of dogs tested at 500 mg/kg.
2. Gastric mucosal damage was produced in rats at 50-200 mg/kg po.
3. Basal gastric acid secretion was inhibited (D-R) in rats at 50-200 mg/kg po. Acetazolamide had no effect at 100 mg/kg po.
4. Colonic propulsive activity was inhibited in mice at 50-200 mg/kg po.
5. Blood pressure and transpulmonary pressure were not affected by 10 mg/kg iv in rats.
6. A transient increase was followed by a prolonged (>24 hr) decrease in MAP after 30 mg/kg ip or 100 mg/kg po in spontaneously hypertensive rats.
7. In anesthetized dogs, 2.5-10 mg/kg iv produced D-R increases in MAP, cardiac output, stroke volume, and left ventricular dP/dt and decreased arterial pH. ECG not affected.
8. In anesthetized dogs, 1-10 mg/kg iv potentiated the pressor effects of epinephrine and phenylephrine.
9. No effects on plasma glucose, insulin, glucagon, or prolactin seen in rats.
10. No *in vitro* effects on lipolysis in rat adipocytes; coronary flow, cardiac rate or contractile force in guinea pig heart; or basal or KCl-stimulated calcium influx in rabbit aorta. Reduced Ca influx stimulated by NE; decreased glucose production in rat hepatocytes; and inhibited antigen-induced contraction of guinea pig lung parenchymal strip.
11. Slight increase in binding of an alpha adrenergic ligand in rat brain homogenate, but no effect on binding of NE or BDZ, or on uptake of 3H-GABA or 3H-norepinephrine.

I) EFFECTS ON EXCITABILITY OF CULTURED FETAL RAT HIPPOCAMPAL NEURONS A500960, Vol. 18).

1. Topiramate (20-100 uM) exerted a direct inhibitory effect on neuronal excitability, reducing burst duration and frequency of action potentials during spontaneous neuronal firing.
2. At 10-100 uM, topiramate blocked depolarization-induced sustained repetitive firing, indicating a possible negative modulatory effect on Na+ and/or Ca++ channels (like PHT and CBZ).
3. Topiramate (10-200 uM) blocked kainate-evoked inward currents, but did not inhibit NMDA-evoked currents.

J) IN VITRO EFFECTS OF TOPIRAMATE IN RAT BRAIN SYNAPTOSOMES (A50612, Vol. 19)

1. Topiramate (10 uM) had no effect on binding of 3H-GABA to the GABAA receptor.
2. Topiramate, PHT, CBZ, acetazolamide, and zonisamide had no effect on resting

membrane potentials at 30 or 100 uM. Partial block of veratrine-induced depolarization was seen with PHT and CBZ but not with topiramate or other drugs at up to 100 uM.

3. No effect on adenosine uptake was seen for topiramate or any drug tested at up to 10 uM.

K) EFFECT OF TOPIRAMATE ON GABAA RECEPTOR CHANNELS (Conducted by A500963, Vol. 19)

Electrophysiological patch-clamp studies with cultured mouse cerebral cortical neurons indicated that topiramate (1-100 uM) enhanced GABA-evoked Cl^- currents. No change in single channel conductances was seen, but the frequency of channel activation was increased. Little effect was seen on open-time duration or duration of burst of channel openings. The results were similar to those for BDZs, which have a very different anticonvulsive profile. No enhancement of GABA-mediated Cl^- flux was observed with acetazolamide (300 uM), suggesting that this action of topiramate is not related to its ability to inhibit carbonic anhydrase.

L) RECEPTOR BINDING AND NEUROTRANSMITTER UPTAKE INHIBITION PROFILES (Conducted A50775, Vol. 19)

Topiramate and MK-801 were tested for *in vitro* radioligand binding and neurotransmitter uptake inhibition in rat and guinea pig brain preparations. Topiramate did not appear to act at any of the binding or uptake sites tested at concentrations up to 10^{-5} M. MK-801 showed high affinity binding to NMDA receptor sites and weakly inhibited NE uptake in rat brain.

M) CHARACTERIZATION OF TOPIRAMATE AS A CARBONIC ANHYDRASE INHIBITOR (Conducted A500961, Vol. 19)

The potency of topiramate as an inhibitor of different isozymes of CA was determined under physiological conditions of CO_2, $HCO3^-$, and H^+ (**Table I.6**).

1. Ki values for inhibition of human erythrocyte CA I and CA II were 90 and 9 uM, respectively.
2. Ki values for rat kidney cytosol (RCA II) and membranes (RCA IV) were 0.08 and 0.18 uM, respectively. Ki value for RCA V in liver microsomes was 18 uM. Ki values of acetazolamide for inhibition of RCA II in kidney cytosol and RCA V in rat liver mitochondria were 0.01 and 0.1 uM, respectively.
3. Ki values for topiramate in rat brain myelin (RCA II), microsomes (RCA IV), and cytosol (RCA II) were 0.05, 0.15, and 0.07 uM, respectively.
4. Conclusions: a) Topiramate appears to be a relatively selective inhibitor of CA isozymes, and a predominant CA isozyme in brain cytosol and myelin (CA II) was sensitive to topiramate. b) Based on topiramate Ki values for human and rat CA II, the drug is 100-times more potent in rats. c) Acetazolamide is a less selective CA inhibitor, and was generally more potent than topiramate; however, the Ki values of topiramate and acetazolamide for inhibition of RCA II were of the same order of magnitude, ie. 0.05-0.08 uM for topiramate vs 0.01 uM for acetazolamide.

Table I.6: Inhibition Constants of Acetazolamide and Topiramate for Rat Carbonic Anhydrase Isozymes

Tissue	Source	Isozyme	Ki (uM) Acetazolamide	Ki (uM) Topiramate
Kidney	membranes	RCA IV	-	0.18
Liver	cytosol	RCA III	>100	>1 mM
Liver	mitochondria	RCA V	0.1	18
Kidney	cytosol	RCA II	0.01	0.08
Brain	myelin	RCA II	-	0.05
Brain	supernatant	RCA II	-	0.07
Brain	microsomes	RCA IV	-	0.15

N) EFFECTS ON GASTRIC ACID SECRETION IN RATS AND DOGS (A500319, Vol. 19)

1. Topiramate inhibited gastric acid secretion after po (ED50 = 188 mg/kg M, 259 mg/kg F) and ip (ED50 = 46 mg/kg M, 156 mg/kg F) administration in pylorus ligated rats. Acetazolamide was less potent after oral (ED50 > 400 mg/kg) and ip (ED50 = 146 mg/kg M, 163 mg/kg F) administration.
2. A significant incidence of gastric lesions was seen after topiramate and acetazolamide in the above rats, but no mucosal irritation at 2, 4, or 18 hr after administration of either drug in nonligated rats.
3. Topiramate inhibited volume of betazole stimulated secretion of gastric juice, acid concentration, and total gastric acid secretion in female chronic fistula dogs (ED50 = 37 mg/kg po). Acetazolamide was more potent against betazole stimulated total acid output in dogs (ED50 = 4.6 mg/kg po).

O) EFFECTS ON RENAL FUNCTION IN RAT (A46970, Vol. 19)

In anesthetized and catheterized rats, topiramate (90 uM/kg (30.5 mg/kg) iv followed by 90 uM /kg/hr continuous infusion) had significant effects on:

1. Renal hemodynamics - decreased MAP and renal blood flow.
2. Urinary water and electrolyte excretion - increased urinary flow rate, Na, K, and HCO3 excretion, and decreased urinary Cl excretion (**Table I.7**).
3. Urinary concentration/dilution - decreased urinary osmolality, increased solute clearance and free water reabsorption.
4. Urinary acidification - decreased arterial blood pH and plasma bicarbonate, increased arterial pCO2; increased urinary pH, pCO2, and bicarbonate, abolished urinary titratable acid excretion, decreased urinary ammonium excretion, reversed net acid excretion.
5. Effects 2-4 were qualitatively similar to, but quantitatively somewhat less pronounced than, effects of an equimolar dose of acetazolamide. Acetazolamide produced no effects on MAP or renal blood flow at an equimolar dose.

Table I.7: Effects of Equimolar Doses (90 uM/kg + 90 uM/kg/hr iv) of Acetazolamide and Topiramate on Urinary Water and Electrolyte Excretion

		Absolute Excretion Rates
V (ul/min)	Control	25.2 ± 3.8
	Acetazolamide	75.3 ± 5.1
	Topiramate	49.5 ± 3.4
$U_{Na}V$ (ueq/min)	Control	3.9 ± 0.6
	Acetazolamide	15.1 ± 1.3
	Topiramate	9.3 ± 0.8
$U_{K}V$ (ueq/min)	Control	2.1 ± 0.1
	Acetazolamide	5.8 ± 0.7
	Topiramate	4.8 ± 0.6
$U_{Cl}V$ (ueq/min)	Control	4.0 ± 0.7
	Acetazolamide	2.6 ± 0.3
	Topiramate	2.7 ± 0.5
$U_{HCO3}V$ (ueq/min)	Control	0.2 ± 0.1
	Acetazolamide	7.5 ± 0.7
	Topiramate	4.4 ± 0.4

P) EFFECTS ON RABBIT INTRAOCULAR PRESSURE (Conducted A50189, Vol. 19)

Carbonic anhydrase inhibitors can reduce intraocular pressure and are used to treat glaucoma. Since topiramate inhibits carbonic anhydrase, a preliminary study was performed to evaluate the ability of topiramate to reduce intraocular pressure in rabbits when administered topically and systemically. The results indicated that topiramate appreciably reduced intraocular pressure when administered iv at doses of 10 or 25 mg/kg but was not active when administered topically. The investigators suggested that further study of topiramate as an ocular hypotensive agent is warranted.

Q) EFFECTS ON BONE LOSS IN NEONATAL MOUSE CALVARIUM CULTURE IN VITRO (A50141, Vol. 19)

Because bone resorption requires carbonic anhydrase, topiramate was tested for possible activity as an inhibitor of bone resorption in cultured neonatal mouse calvarial tissue. Topiramate inhibited calcitriol-stimulated bone resorption at 10^{-3} M but not at lower concentrations. The potency was considered insufficient to merit further evaluation as a drug for treating osteoporosis and related disorders.

R) INTERACTION WITH OTHER ANTICONVULSANTS (A47239, Vol. 17)

Male SW mice were given oral doses of topiramate in combination with other AEDS (DPH, PHB, CBZ) in fixed ratios of their MES ED50s (0.75/0.25, 0.5/0.5, 0.25/0.75). 3-5 doses of each combination were used in order to construct an anticonvulsant dose-response curve for each fixed ratio. The determination of possible interactions was made by calculating ED50 values and testing for synergism and parallelism using maximum likelihood techniques. Based on an analysis of the isobolograms generated for the three different ratios of topiramate and phenytoin in mice, the anticonvulsant activity of these drugs was additive. When topiramate was combined with carbamazepine or phenobarbital, the anticonvulsant activity was slightly but statistically significantly synergistic at the 50/50 ratio.

II. ADME

A) ABSORPTION AND PHARMACOKINETICS

1. BIOAVAILABILITY AND PK IN MALE RATS (A47315 Vol. 41)

Single iv (15 mg/kg) or oral (30 mg/kg) doses were administered to two groups of six male rats (Sprague-Dawley) per route. Serial samples were collected by orbital sinus puncture at 5 or 6 timepoints for each group; timepoints were selected so that data from the two groups per route could be combined into one series with each group providing alternate timepoints. Drug concentrations were determined by a capillary GC method. Results are shown in **Table II.1**.

Table II.1: Mean Pharmacokinetic Parameters following IV (15 mg/kg) or PO (30 mg/kg) Administration of Topiramate to Sprague-Dawley Rats (6/sex)

Parameters	Male		Female	
	IV	PO	IV	PO
Cmax (ug/ml)	17.8	27.6	23.1	33.8
tmax (hr)	-	0.5	-	0.9
t1/2 (hr)	2.2	2.6	5.2	4.7
AUC (ug·hr/ml)	68	138	163	319
Cl/F(ml/min/kg)	3.7	3.6	1.5	1.6
F	-	1.01	-	0.98

2. BIOAVAILABILITY AND PK IN FEMALE RATS (A500753, Vol. 42)

Single iv (15 mg/kg) or oral (30 mg/kg) doses were administered to groups of six female rats (Sprague-Dawley) per route. Serial samples were collected by retro-orbital puncture at 8 timepoints per group.

IV: The mean extrapolated C0 was 23.1 ug/ml, t1/2 was 5.18 hr, AUC (0-30 hr) was 163 ug·hr/ml, Cl was 92 ml/hr/kg, and Vd was 0.690 L/kg.

Oral: A mean Cmax of 33.8 ug/ml was measured at 0.9 hr, t1/2 was 4.70 hr, AUC was 319 ug·hr/ml, Cl/F was 94 ml/hr/kg, and absolute bioavailability was 98%.

3. EFFECT OF DOSE ON PK IN MALE AND FEMALE RATS (233277:2, Vol. 42)

10 Wistar rats/sex received a 10 mg/kg oral solution dose of topiramate followed 2 weeks later by a 300 mg/kg oral suspension dose. Blood samples were obtained from the orbital sinus at specified time points through 24 hr post-dosing. Results are shown in **Table II.2**.

Table II.2: Pharmacokinetic Parameters of Topiramate in Wistar Rats Following Single Oral Dose of Topiramate as a Solution (10 mg/kg) or Suspension (300 mg/kg)

	Male		Female	
Parameter	10 mg/kg	300 mg/kg	10 mg/kg	300 mg/kg
AUC (ug·hr/ml)	31.7	1070.0	102.0	2703.5
NAUC	3.17	3.57	10.2	9.01
Clearance/F (ml/min/kg)	5.27	4.67	1.63	1.85

AUC: Area under the serum concentration vs time curve, 0-24 hr.
NAUC: Dose normalized AUC.
Clearance/F: Calculated as Dose/AUC.

At each dose, AUC values for female rats were about 2.5 times those for males, and clearance was correspondingly lower in females than in males. Dose-normalized AUC values were equivalent within sex, indicating dose-proportional absorption.

4. EFFECT OF REPEATED DOSING ON PK IN MALE & FEMALE RATS (A500923, Vol. 42)

6/sex/group (Sprague-Dawley) were administered either a single oral gavage dose (30 mg/kg) or 8 daily oral doses (30 mg/kg/day). Blood samples were taken from the orbital sinus at 0.5, 1, 2, 4, 6, and 8 hr post-dose on either day 1 (single does group) or day 8 (multiple dose group). Results are shown in **Table II.3**.

Table II.3: Pharmacokinetic Parameters (mean ± SD) of Topiramate (30 mg/kg) in Rats Following Single or Multiple Oral Administration

	Male		Female	
Parameter	Day 1	Day 8	Day 1	Day 8
Cmax	11.5 ± 1.6	14.1 ± 3.1	18.9 ± 2.0	22.2 ± 4.3
Tmax	0.80 ± 0.27	0.75 ± 0.27	2.0 ± 0.00	1.33 ± 0.52
AUC (ug·hr/ml)	48.3 ± 2.8	69.0 ± 11.9	257.2 ± 34.4	268.2 ± 21.0
Clearance/F (ml/min/kg)	10.39 ± 0.62	7.45 ± 1.41	1.97 ± 0.25	1.88 ± 0.15
t1/2 (h)	2.35 ± 0.50	2.39 ± 0.31	5.99 ± 1.10	6.80 ± 2.81

No significant within sex differences were found between values obtained on days 1 and 8 for AUC, Cmax, and t1/2, indicating no accumulation or autoinduction.

5. PK IN FEMALE RABBITS FOLLOWING SINGLE OR MULTIPLE DOSING (A500913, Vol. 42)

Eight female New Zealand White rabbits were administered a single oral gavage dose (60 mg/kg) of topiramate, and serial blood samples were taken up to 24 hr post-dosing. Rabbits were then dosed for another 13 days (60 mg/kg/day) and serial samples taken after dosing on day 14. Results are shown in **Table II.4**.

Table II.4: Pharmacokinetic Parameters (mean ± SD) of Topiramate (60 mg/kg) in Female Rabbits After Single or Multiple Oral Administration

Parameter	Day 1	Day 14
Cmax	39.5 ± 7.9	39.1 ± 6.0
Tmax	1.72 ± 1.05	1.25 ± 0.52
AUC (ug·hr/ml)	212 ± 28	201 ± 42
CL/F (ml/min/kg)	4.80 ± 0.73	5.19 ± 1.21
t1/2 (h)	2.85 ± 0.40	2.49 ± 0.37

No differences were found between values on days 1 and 14 for AUC, Cmax, and t1/2, indicating no accumulation or autoinduction.

6. PK IN MALE DOGS AFTER SINGLE IV OR ORAL DOSE (A500927, Vol. 42)

Single iv (10 mg/kg) or oral (300 mg/kg) doses were administered to 1 male dog per route. Serial blood samples were then collected for 24 hr. Plasma topiramate concentrations were measured by capillary GC assay.

IV: A mean Cmax of 18.9 ug/ml was observed at 5 min (1st timepoint), the t1/2 was 6.5 hr, and the AUC (0-24 hr) was 149.1 ug·hr/ml.

Oral: A mean Cmax of 201.1 ug/ml was measured at 4 hr, the t1/2 was 4.70 hr, and the AUC was 3249 ug·hr/ml. Absolute bioavailability was 73%.

7. PK IN DOGS AFTER SINGLE OR MULTIPLE ORAL DOSES IN GELATIN CAPSULES (A49953; Vol. 42)

Three groups of 2/sex were given single doses of 10, 40, or 150 mg/kg, and serial plasma samples were collected up to 78 hr post-dosing. Two weeks after the single dose a multiple dose regimen was begun (10, 40, and 150 mg/kg/day for 15 days), with plasma samples collected over 96 hr after last dose. Only combined sex PK values were reported. (See **Table II.5**).

Linearity between dose and Cmax or AUC was observed after single and multiple dosing. Cmax values increased dose-proportionally after single and multiple dosing. After a single dose, LD and MD AUCs were dose-proportional, but HD AUCs were greater than proportional. After multiple dosing, LD and HD AUCs were dose-proportional, but MD AUCs were less than proportional. Vd decreased with increasing dose after both single and

multiple dosing, which was attributed to binding of drug to erythrocytes (ie, RBCs acting as deep compartment). This might also explain the longer t1/2s at the LD after single and multiple dosing. No biologically significant differences in PK parameters were seen after single and multiple dosing at each dose.

Table II.5: Pharmacokinetic Parameters (mean ± SD) of Topiramate in Male and Female Beagle Dogs After Single or Multiple Oral Administration

Dose Regimen and Parameters	Dose Group (mg/kg)		
	10	40	150
Single Dose			
Cmax (ug/ml)	9.2 (1.8)	45.4 (10.8)	137.7 (47.8)
Tmax (h)	2.4 (2.5)	1.4 (0.25)	3.9 (1.8)
AUC (ug·h/ml)	50.7 (6.6)	190.0 (25.3)	1131.0 (296.0)
t1/2 (h)	3.7 (0.22)	2.6 (0.33)	2.9 (0.63)
Cl/F(ml/min/kg)	3.4 (0.45)	3.6 (0.46)	2.4 (0.81)
Vd (L/kg)	1.09 (0.10)	0.79 (0.11)	0.62 (0.37)
Multiple Dose			
Cmax (ug/ml)	10.3 (2.1)	43.5 (9.2)	145.2 (20.1)
Tmax (h)	2.4 (1.1)	1.1 (0.31)	1.6 (0.25)
AUC (ug·h/ml)	54.4 (6.0)	161.4 (25.4)	858.1 (154.6)
t1/2 (h)	3.8 (0.65)	2.0 (0.11)	2.2 (0.13)
Cl/F(ml/min/kg)	3.1 (0.35)	4.2 (0.67)	3.0 (0.49)
Vd (L/kg)	1.02 (0.11)	0.73 (0.12)	0.56 (0.07)

B) TISSUE DISTRIBUTION

1. TISSUE DISTRIBUTION OF RADIOACTIVITY IN RATS (A500283,Vol. 44)

16 male Wistar rats were given single 10 mg/kg oral dose of 14C-topiramate, and 4/time-point were sacrificed at 1, 6, 24, and 48 hr. Peak blood and plasma levels of radioactivity measured at 1 hr were 21 and 12 ug-equiv/ml, respectively. At 1 hr, parent accounted for 60% of total radioactivity, and the highest levels of radioactivity were found in the gi tract, liver, kidneys, thyroid, and pituitary. Within 48 hr of dosing 104% of the dose was excreted in urine (67%) and feces (37%).

2. TISSUE DISTRIBUTION IN PREGNANT AND NONPREGNANT FEMALE RATS (A500331, Vol. 44)

A single oral dose of 14C-topiramate (20 mg/kg) was given to 16 pregnant Sprague-Dawley rats on day 11 of gestation and to 4 nongravid females. Concentrations of radioactivity were determined in groups of 4 pregnant animals at 1, 6, 24, and 48 hr. Levels were determined in nonpregnant rats at 6 hr only.

Highest levels were found in pregnant rats at 1 or 6 hr in blood (20.8 ug-equiv/g), gi tract (577), liver (19), and kidney (17.5). Concentrations in the fetus were similar to plasma

levels and declined in parallel with plasma. Most (80-90%) of total radioactivity in plasma, liver, kidney, and fetus during 48 hr was unchanged parent. (Higher % unchanged in females than males, ie, sex difference in metabolism.) Approximately 68% of administered dose was eliminated in urine by 48 hr. Tissue levels in nonpregnant females were similar to those in pregnant rats at 6 hr. Concentrations in gravid females at 24 and 48 hr were much higher than those previously found in male rats.

3. QUANTITATIVE WHOLE-BODY AUTORADIOGRAPHY AFTER ORAL DOSE IN RATS (A505022, Vol. 44)

4 Wistar rats/sex were given 10 mg/kg 14C-topiramate by gavage, and 1/sex sacrificed at 1, 6, 24, and 48 hr. Radioactivity as rapidly and extensively absorbed and distributed in both sexes, but was eliminated from males at a much higher rate, with higher levels remaining in females at 48 hr. At 1 hr, levels of radioactivity were comparable between sexes, with highest levels (10-50 ug-equiv/g) in gi tract, liver, kidneys, blood, lung, myocardium, spleen, salivary glands, and adrenals. Brain levels of 4.6 and 6.4 ug-equiv/gm were measured in males and females, respectively, at 1 hr. Levels declined rapidly in males such that at 24 hr most tissues contained <1 ug equiv/gm, and levels in all but a few tissues were not quantifiable at 48 hr. In females, levels were at least twice those in males at 6 hr, and the difference was even more marked at 24 and 48 hr. Although not quantified, moderate levels of radioactivity were noted in the stomach mucosa at most kill times.

C) METABOLISM AND EXCRETION

1. METABOLISM AND EXCRETION IN MICE (A500912, Vol. 45)

45 CD-1 mice/sex were given a single oral dose of 14C-topiramate (300 mg/kg), and urine and feces were collected from 15/sex for 5 days. Blood was collected from 6/sex at 0.5, 1, 4, 8, and 24 hr.

A total of 99.7% of administered dose was recovered in urine (88.4%), feces (9.8%), and cage wash (1.5%) over 5 days. Representative samples of plasma, RBCs (0.5-8 hr), urine (0-24 hr pool, 83%), and fecal extract (0-24 hr pool, 6%) were purified for metabolite profiling, isolation, and identification. Unchanged topiramate and four metabolites were identified (**Table II.6**).

Unchanged topiramate accounted for 87%, 88%, 61%, and 31% of radioactivity in plasma (0.5 hr), RBCs (0.5-8 hr), urine (0-24 hr), and fecal extract (0-24 hr) pools, respectively. Four metabolites were identified, which are thought to be formed by the following proposed metabolic pathways (**Figure II.1**): A) hydroxylation at 7- or 8-methyl followed by rearrangement to yield M-1, B) hydroxylation at 10-methyl to yield M-2, C) hydrolysis at 2,3-O-isopropylidene to produce a diol (M-4), and E) cleavage at the sulfamate group to give M-6. Pathway C appeared most important; M-4 was the only major metabolite identified. The minor metabolite M-2 showed some anticonvulsant activity, but was less potent than topiramate.

Table II.6: Percent of Dose and Sample Radioactivity Accounted for by Topiramate and Its Metabolites in Mice

	Percent 14C of Dose		Percent 14C in Sample	
	Urine (0-24 h)	Feces (0-24 h)		Plasma (0.5-5 h Pool)
Total Sample	83.4	6.1		
Topiramate	51	2	Topiramate	87
M-1	<2	<2	M-1	<5
M-2	<2	<2	M-2	<5
M-4	17	2	M-4	8
M-6	ND	<2	M-6	ND

ND = None Detected

2. METABOLISM AND EXCRETION IN RATS (233528:2, Vol. 45)

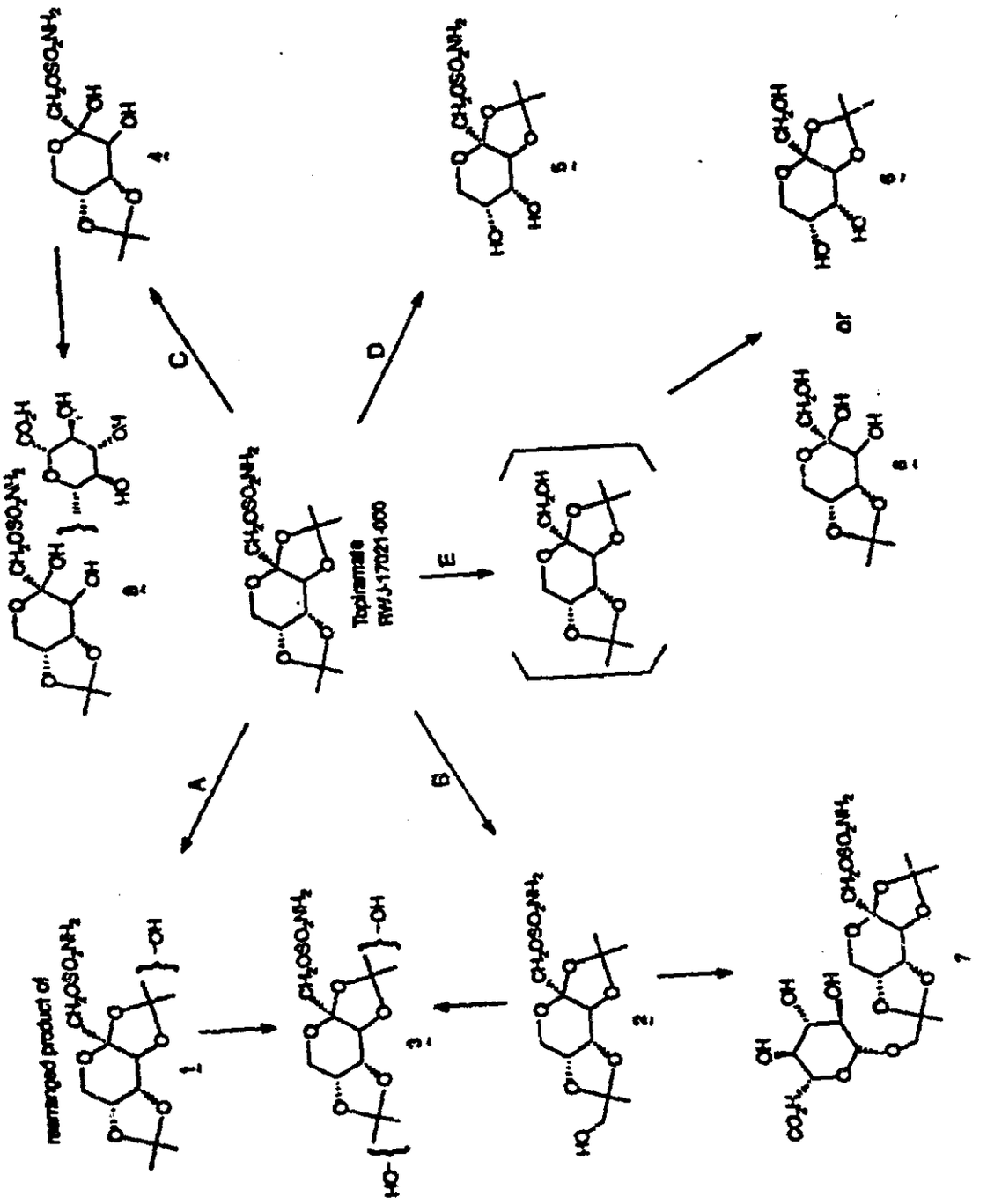
Plasma, urine, and feces were analyzed for total radioactivity and topiramate after administration of single oral doses (90 mg/kg) of 14C-topiramate to rats (4/sex for blood, 4/sex for urine and feces).

The major component of total radioactivity in plasma was topiramate: 99% at 1.5 hr after dosing, 74% at 7 hr. Females excreted a significantly greater fraction of dose in urine (88%) than males (70%). Unchanged drug accounted for a greater fraction of total urinary radioactivity in females (92%) than in males (60%). Fecal excretion of total radioactivity was greater in males (27% of dose) than in females (6%). Less than 1.5% of fecal radioactivity was topiramate in both sexes. (See **Table II.7**).

Table II.7: Total Radioactivity excreted in Urine and Feces (0-96 h) from Male and Female Rats following a Single Oral Dose of Topiramate (90 mg/kg)

Male Rats				Female Rats			
Rat No.	Urine	Feces	Total	Rat No.	Urine	Feces	Total
1	62.7	31.8	94.5	1	79.8	9.5	89.3
2	82.3	24.5	106.8	2	92.2	2.9	95.1
3	59.0	26.6	85.6	3	86.3	7.3	93.6
4	73.5	23.5	97.0	4	93.2	6.0	99.2
Mean (SD)	69.4 (10.59)	26.6 (3.70)	96.0 (8.72)		87.9 (6.18)	6.4 (2.76)	94.2 (4.09)

Figure II.1: Metabolic Pathways of Topiramate



3. METABOLISM IN RAT AND DOG (A500709, Vol. 45)

Plasma, urine, and fecal samples were obtained from rats (90 mg/kg) and dogs (40 mg/kg), and metabolites were isolated and identified. Six metabolites were identified in rats and four in dogs. Topiramate and identified metabolites accounted for >78% of total in urine, >92% in plasma, and >50 in fecal extracts in both species. Unchanged topiramate accounted for more of dose in female than in male rat urine and fecal samples, as noted previously (**Table II.8**).

Five metabolic pathways are proposed for both species (**Figure II.1**): A) hydroxylation at 7- or 8-methyl of isopropylidene followed by rearrangement (M-1), B) hydroxylation at 10-methyl of isopropylidene (M-2), C) hydrolysis at 2,3-Ω-isopropylidene (M-4), D) hydrolysis at 4,5-Ω-isopropylidene (M-5), and E) cleavage at the sulfamate group (M-6). M-3 is thought to be a product of a second hydroxylation of either M-1 or M-2. In both species, hydroxylation at the isopropylidene (paths A and B) produced the two OH-topiramates, (M-1 & 2) in significant quantities (9-20% of sample) and appear to constitute the major metabolic pathway. Pathway C is also quantitatively important, resulting in metabolite 4, a major metabolite in urine and feces in both species (10-20% of sample). Pathways D and E were less important; the two minor metabolites formed represented <5% of total radioactivity in any sample pool. Enzyme hydrolysis of dog urine samples did not reveal the presence of significant amounts of conjugated metabolites.

Table II.8: Percent of Total 14C in Sample for Topiramate and its Metabolites

Sample	Plasma			Urine (0-24 h)			Feces (0-24 h)		
Species	Rat		Dog	Rat		Dog	Rat		Dog
Sex	M	F	M&F	M	F	M&F	M	F	M&F
% Dose	NA	NA	NA	63.2	78.7	82.7	16.7	2.9	4.6
Topiramate	87.7	90.8	91	47.4	85.5	28.1	4.6	31.9	25.0
M-1	+	+	+	9.0	+	14.3	12.5	12.1	19.4
M-2	+	+	+	+	+	12.4	9.8	12.2	20.0
M-3	-	-	-	-	-	-	+	+	-
M-4	+	+	+	11.6	+	20.0	17.8	15.5	16.0
M-5	-	-	-	+	+	+	+	+	-
M-6	-	-	-	+	-	-	-	-	-
Total % 14C in Sample identified	92	94	95	80	90	78	50	72	80

4. BILIARY AND MILK SECRETION IN RATS (A500943, Vol. 45)

Metabolite profiles were determined in bile and milk after administration of single oral doses of 14C-topiramate to bile duct-cannulated (6/sex) or lactating (12 female) Wistar rats. 37.5% and 5.3% of the radioactive dose was excreted in 24-hr bile in male and female rats, respectively (**Table II.9**). In males, metabolites accounted for >96% of bile radioactivity over 24 hr; in females, metabolites accounted for about 60-70% of radioactivity in bile. Enzyme hydrolysis increased the % intact topiramate in selected samples from <3%

to >19% in males and from <40% to >43% in females and decreased % polar metabolites, indicating that glucuronide conjugates were present in major quantities. Three metabolites were identified (**Figure II.1**): metabolites 2 and 4 in major quantities, and metabolite 1 in minor quantities.

Table II.9:Percent of Dose and Sample Radioactivity Accounted for by Topiramate and its Metabolites in Glusulase-treated Rat Bile Pools

Percent 14C of Dose			Percent 14C in Sample	
	Male (0-24 h)	Female (0-24 h)	Male (0-24 h)	Female (0-24 h)
Total Sample	37.5	5.3		
Topiramate	11	3	28	65
M-1	<2	<1	<5	<5
M-2	12	1	32	20
M-4	10	<1	27	11

About 1.5% of the radioactive dose was recovered in milk over 24 hr postdose. No significant amounts of metabolites (<0.2% radioactive dose) were present in milk from 1 to 8 hr after dosing. The Cmax of total radioactivity in milk was 6 ug-equiv/ml at the Tmax of 2 hr. The ratio of milk/plasma concentrations and AUC ranged from 0.8 to 1.1 indicating, a close relationship between plasma and milk. The elimination half-life in milk (6.4 hr) was comparable to that in plasma (5.8 hr) but shorter than that in blood (15.9 hr). Cmax values of total radioactivity in plasma and blood were 7.16 and 15.9 ug-equiv/ml, respectively.

5. METABOLISM AND EXCRETION IN FEMALE RABBIT (A500911, Vol. 45)

A single 60 mg/kg oral dose of 14C-topiramate was given to 6 female New Zealand White rabbits, and plasma, blood, urine, and fecal samples were analyzed for total radioactivity and topiramate. Representative samples were purified for metabolite identification.

Total radioactivity in plasma at 2 hr was 50 ug-equiv/ml. Total percentages of the radioactive dose excreted in urine and feces were 79% and 10.2%, respectively. About 93% of total urinary radioactivity and 68% of total fecal radioactivity was excreted within 24 hr. The inactive diol (metabolite 4) and the active OH-topiramate (metabolite 2), were major and minor metabolites, respectively (see structures in **Figure II.1**).

6. ABSORPTION, EXCRETION, AND BIOTRANSFORMATION FOLLOWING A SINGLE ORAL DOSE IN MALE AND FEMALE DOGS (233531:2, Vol 42)

Two dogs/sex were given single oral doses (40 mg/kg) of 14C-labeled topiramate by gavage. Serial blood samples, urine, and feces were collected up to 96 hr after dosing.

There were no apparent sex differences in topiramate pharmacokinetics (but Ns small). Cmax values measured at 0.6-0.8 hr averaged about 50 ug/ml. Vd averaged 0.57 L/kg, close to total body water. Drug plasma elimination half-life averaged 3.3 hr and total plasma radioactivity declined at a similar rate. Clearance averaged 2 ml/min/kg. During the

first 24 hr, about 25% of administered dose was excreted unchanged in urine in both sexes and only 1-2% in feces. Females excreted somewhat more radiolabel in urine than males (99 vs 82%) and correspondingly less in feces (3 vs 9%) over 96 hr. TLC analysis revealed the presence of at least 3 nonhydrolyzable metabolites in urine and feces from both sexes. (Results summarized in **Table II.10**).

Table II.10: Plasma, Urine and Fecal Concentrations of Topiramate and Total Radioactivity following Administration of a Single Oral Dose (40 mg/kg) of 14C-Topiramate to Dogs (2/sex)

Plasma Pharmacokinetics (unchanged topiramate)	Male	Female
Tmax (h)	0.6	0.8
Cmax (ug/ml)	49.7	59.2
[% of total 14C]	100	92
C24 h (ug/ml)	0.4	0.5
[% of total 14C]	39	24
AUC (ug·h/ml)	334.9	350.9
t1/2 (h)	3.3	3.4
CL/F (ml/min/kg)	2.0	1.9
Vd (L/kg)	0.57	0.57
Excretion (% of 14C Dose)	**Male**	**Female**
Urine - Topiramate	25.4	26.9
Total 14C (0-24 h)	73.9	91.5
Total 14C (0-96 h)	81.5	98.7
Feces - Topiramate	2.2	<1.0
Total 14C (0-24 h)	7.3	2.1
Total 14C (0-96 h)	8.9	2.8
Recovery Total 14C	90.2	101.6

D) PROTEIN BINDING

1. IN VITRO PROTEIN BINDING (A44651; Vol. 43)

Binding to plasma proteins and erythrocytes was assessed in several species by equilibrium dialysis. Low protein binding was found in mouse (10-16%), rat (12-15%), rabbit (13-16%), dog (8-13%), monkey (2-14%), and human plasma (11-17%) at concentrations ranging from 1-300 ug/ml. Evidence of saturable protein binding was seen in mouse, monkey, and human. A high affinity, low capacity erythrocyte binding site was identified in all species tested.

E) ENZYME INDUCTION

1. EFFECTS OF ORAL ADMINISTRATION OF TOPIRAMATE FOR TWO WEEKS ON HEPATIC MICROSOMAL ENZYMES IN RATS (A50617, Vol. 45)

Sprague-Dawley rats (3/sex) were given daily oral doses (750 mg/kg) of topiramate for 2 weeks and selected indicators of microsomal enzyme activity were assessed relative to a vehicle control group (3/sex).

Topiramate treatment for 2 weeks resulted in increases (116%-361%) in several measures of drug metabolizing activity. A sex difference was seen in the effect on benzo(a)pyrene hydroxylase activity, which was increased (3.5X) only in females.

2. EFFECTS OF ORAL ADMINISTRATION FOR NINE MONTHS ON MICROSOMAL ENZYMES IN RATS (233292:2; Vol. 45)

3/sex/group were dosed with 0, 10, 55, or 300 mg/kg/day (diet) for 9 months, and the following indicators of hepatic microsomal enzyme activity were determined: protein content, cytochrome P450 content (P450), 7-ethoxycoumarin O-deethylase (ECOD) activity, erthoxyresorufin O-deethylase (EROD) activity, and acetaminophen glucuronyltransferase (AGT) activity. EROD activity specifically reflects induction of isozymes from the cytochrome P450IA subfamily. ECOD reflects induction of a broader spectrum of isozymes, including P450IA and IIB (phenobarbital-inducible) subfamilies.

Dose-related increases (129-443%) were seen for most measures, particularly EROD and ECOD activities. In the ECOD assay, enzyme activity increased more in males than in female, while the reverse was true in the EROD assay. Effects on P450 content and AGT activity were less pronounced and similar between sexes at the HD. Protein content only increased in HD females. Comparison of topiramate effects with historical data for phenobarbital and beta-naphthaflavone is shown in **Table II.11**.

Table II.11: Comparison of the Abilities of Topiramate, β-Naphthaflavone, and Phenobarbital to Induce Hepatic Drug Metabolizing Enzymes in Wistar Rats

			% Control			
Sex	Compound	Dose[a]	Protein	P450	ECOD	EROD
M	Topiramate	300[b]	131 ± 9	129 ± 8	352 ± 24	170 ± 27
M	Phenobarbital	75[c]	126 ± 16	188 ± 25	350 ± 64	167 ± 26
F	Topiramate	300[b]	114 ± 13	140 ± 10	220 ± 57	458 ± 30
F	β-Naphthaflavone	80[c]	120 ± 7	201 ± 13	880 ± 70	19776 ± 5291
F	Phenobarbital	75[c]	121 ± 5	167 ± 25	341 ± 68	86 ± 18

[a] mg/kg/day
[b] Animals dosed daily for 9 month (n = 3/group)
[c] Animals dosed daily for 3 days (n = 3/group)

F) DRUG INTERACTIONS

1. TOPIRAMATE METABOLISM AND EXCRETION AFTER PRETREATMENT WITH OR COADMINISTRATION OF PHENYTOIN IN FEMALE RATS (A500681, Vol. 45)

Eighteen female rats (Sprague-Dawley, 6/group) were assigned to 3 treatment groups: A) pretreatment with phenytoin vehicle for 7 days before receiving single 60 mg/kg oral dose of 14C-topiramate, B) simultaneous single oral doses of 14C-topiramate (60 mg/kg) and phenytoin (100 mg/kg), and C) pretreatment with oral phenytoin (100 mg/kg/day) for 7 days prior to single oral dose of 14C-topiramate (60 mg/kg). Quantitative urine (0-4, 4-8, 8-24 hr) and fecal (0-24 hr) collections were made in 2/group, and serial blood was collected from 4/group (0.5-8 hr).

Renal elimination of radioactivity was decreased or delayed by phenytoin co-administration, ie, urine recovery 24-27% was lower in group B than in groups A and C. Unchanged topiramate accounted for 90-95% of radioactivity in plasma over 8 hr and 83-96% of radioactivity in urine over 24 hr in groups A and B. In group C, unchanged topiramate accounted for 79-84% of radioactivity in plasma (0.5-8 hr) and 47-53% of radioactivity in urine (0-24 hr). AUC for total radioactivity was 34% lower, and plasma, urinary, and fecal metabolites were 2-3 fold higher in group C than in groups A and B, indicating induction of topiramate metabolism by phenytoin in female rats.

2. PHARMACOKINETIC INTERACTIONS BETWEEN PHENYTOIN, CARBAMAZEPINE, AND TOPIRAMATE IN MALE RATS (A500924, Vol. 46)

Five groups of male Sprague-Dawley rats (9/group) were given daily oral doses of PHY (100 mg/kg, 2 groups), CBZ (25 mg/kg, 2 groups), or vehicle (1 group) for 8 days. On day 8, 3 groups (PHY, CBZ, VEH) were also given single oral dose of topiramate (30 mg/kg). Serial blood samples were then collected from all 5 groups, and PK comparisons made for: 1) PHY, with and without topiramate, 2) CBZ, with and without topiramate, and 3) topiramate, alone and with PHY or CBZ.

There were no differences in PHY and CBZ PK parameters due to addition of topiramate. The only effect of CBZ pretreatment and coadministration on topiramate PK parameters was a decrease (30%) in Cmax. A threefold increase in topiramate clearance and a corresponding decrease in AUC was observed with pretreatment and coadministration of PHY, again indicating induction.

3. TOPIRAMATE METABOLITE PROFILES AFTER PRETREATMENT WITH PHENYTOIN, CARBAMAZEPINE, OR TOPIRAMATE (A500838, Vol. 46)

Four groups of male Sprague-Dawley rats (2-4/group) were given daily oral doses of PHY (100 mg/kg), CBZ (25 mg/kg), topiramate (30 mg/kg), or vehicle for 7 days. On day 8, rats in each group were given single oral dose of 14C-topiramate (30 mg/kg) and metabolic profiles were analyzed in plasma, brain, and liver.

In the vehicle group, unchanged topiramate accounted for 83, 96, and 72% of radioactivity in plasma, brain, and liver, respectively. In the phenytoin and topiramate groups, % parent decreased (5-26%) and metabolites increased (1.6-2.8X) compared to vehicle. Pretreatment with carbamazepine had no effect on the biotransformation of topiramate.

4. EFFECT OF PROBENECID OR RENAL IMPAIRMENT ON TOPIRAMATE PK IN FEMALE RATS (A500934, Vol. 46)

Rats received the following treatments: topiramate alone (60 mg/kg po, N=6), topiramate (60 mg/kg po) with ip probenecid (100 mg/kg) at 0, 4, and 8 hr (N=6), 14C-topiramate (60 mg/kg po) alone (N=3) or with probenecid (100 mg/kg ip at 0, 4, & 8 hr, N=3), and topiramate (60 mg/kg po) and methoxy-inulin 5 days after induction of renal impairment with 0.5, 1, 3, or 5 mg/kg sc uranyl nitrate (6/dose). Plasma and urine were collected for 24 hr after topiramate. (Topiramate metabolism in female rats was said to most closely approximate human metabolism.)

Co-administration of probenecid resulted in a significantly lower Cmax (39%) and AUC (45%) and a correspondingly higher Cl/F (111%). Elimination half-life was unaffected. Renal clearance was increased (55%). Probenecid did not alter the absorption or metabolism of radiolabeled topiramate. Thus, results are consistent with an increase in renal clearance due to inhibition of carrier-mediated reabsorption.

In renal impaired rats, effects on topiramate were variable due to variable degrees of renal damage. In mild cases, clearance appeared to increase, possibly due to renal tubule transport damage. Increasing damage resulted in compensatory reduction in GFR, so that little net change in topiramate CL was seen. Only in severe renal impairment (GFR <20% normal) was clearance decreased.

5. EFFECT OF ETHANOL ON TOPIRAMATE PK IN MALE RATS (A500933, Vol. 46)

Twenty six male Wistar rats received 1 of 3 treatments: 1) single topiramate dose (200 mg/kg po) followed 1 hr later by single dose of vehicle (N=6), 2) topiramate (200 mg/kg po) followed 1 hr later by single dose of ethanol (2.4 g/kg ip, N=10)), or 3) vehicle followed by ethanol (2.4 g/kg ip, N=10). One, 6, and 5 rats from groups 1, 2, and 3, respectively, died.

No changes in topiramate PK parameters were produced by ethanol co-administration. The duration of ethanol-induced narcosis was significantly increased by topiramate co-administration.

6. HUMAN LIVER MICROSOMES (A505011; Vol. 45)

The inhibitory spectrum of topiramate was characterized in human liver microsomes. Topiramate (up to 1mM) did not inhibit reactions catalyzed by CYP1A2, 2A6, 2C9, 2D6, 2E1. or 3A4. Inhibition of CYP2Cmeph was indicated by decreased rates of formation of 4'-hydroxymephenytoin from S-mephenytoin in microsomal incubations containing topiramate (300-900 uM). Since CYP2Cmeph is thought to contribute to phenytoin metabolism (in addition to CYP2C9), this may explain the effect reported in a clinical interaction study, ie, phenytoin clearance decreased in a few subjects when topiramate was introduced to treatment regimens.

Attempts to determine the P450 isoforms catalyzing formation of the primary topiramate metabolite were unsuccessful. Topiramate metabolites were not detected after 14C-topiramate was incubated with human liver microsomes.

III. TOXICOLOGY

A) ACUTE ORAL TOXICITY IN MICE (A47161, Vol. 20)

Deaths were seen at 1-6 days postdosing, the majority within 1-2 days. Males appeared more sensitive based on the mortality dose-response. Clinical signs included decreased activity, ataxia, ptosis, loss of righting reflex, and clonic convulsions (at > 2250 mg/kg).

Dose (mg/kg)	M F	1000 1000	1500 1500	2250 2250	- 2750	3375 3375
Mortality	M F	0/2 0/2	0/5 0/2	2/5 0/5	- 1/5	5/5 5/5
Estimated LD50 (95% Confidence Limits)	M F	2338 (1585-3752) mg/kg 2915 (2480-3844) mg/kg				
Observed Maximum Non-Lethal Dose	M F	>1500<2250 mg/kg >2250<2750 mg/kg				

B) ACUTE ORAL TOXICITY IN RATS (A47163, Vol. 20)

Deaths were seen at 2-4 days postdosing. Males were less sensitive based on the mortality dose-response. Clinical signs included decreased activity, ataxia, ptosis, loss of righting reflex (at > 1500 mg/kg), tremors, and convulsions (at > 1500 mg/kg). Congestion of meninges was found at postmorten examination (probably secondary to seizures).

Dose (mg/kg)	M F	1500 1500	2250 2250	— 2750	3375 3375	4220 —
Mortality	M F	0/2 0/2	1/5 1/5	— 5/5	2/5 5/5	3/5 —
Estimated LD50	M F	3745 mg/kg 2436 mg/kg				
Observed Maximum Non-Lethal Dose	M F	1298 mg/kg 1716 mg/kg				

C) ACUTE ORAL TOXICITY IN DOGS (A47034, Vol. 20)

Two groups of beagle dogs received single oral doses of 270 (2/sex) or 400 mg/kg (1/sex). No deaths occurred. Clinical signs were seen in males only at both doses, and included ataxia, decreased activity, and salivation. In addition, one HD male had intermittent clonic convulsions, tremors, and delayed emesis. All signs except emesis (seen at 8-24 hr) were seen within 5-8 hr post-treatment, and all signs had remitted by 24 hr.

D) 3-MONTH ORAL TOXICITY IN RATS (A47056, conducted by GLP; Vol. 21)

1. Treatment

15/sex/group were dosed with 0, 10, 90, or 750 mg/kg/day, by gavage.
Strain: Sprague-Dawley [Crl:CD(SD)BR]
Drug lot #: 8420060

2. Mortality

One HD female sacrificed moribund during week 13 was the only treatment-related death.

3. Clinical signs

Ataxia, salivation, dyspnea, lacrimation, poor grooming, and thin and hunched appearance were observed with a dose-dependent incidence.

4. Body weight and food consumption

a) BW gain was decreased in MD (15%) and HD (25%) females compared to C. Final mean BW was lower in MD females (9%) and HD males (5%) and females (13%) compared to C.
b) No differences in food consumption.

5. Ophthalmoscopic examination

No treatment effects.

6. Hematology (once pretest and in weeks 2, 5, & 13 in 10/sex/group)

RBCs were slightly increased (week 2) and MCHC slightly decreased (week 13) in HD males. This and some of the clinical chemistry findings (increases in protein, albumin, globulin, & BUN) were attributed to hemoconcentration, secondary to diuresis.

7. Clinical Chemistry (once pretest and during weeks 2, 5, & 13 in 10/sex/group)

a) Potassium decreased in HD females (10% below C).
b) Total protein, albumin, and globulin slightly increased in HD males (5-10%) and females (15%).
c) Bilirubin increased in HD males and females (about 50% in both sexes).
d) BUN increased in HD males and females (about 50% in both sexes).
e) Cholesterol and triglycerides increased in HD females (both by about 100%); triglycerides decreased in HD males (50%).
f) AST decreased (20%) in HD females.

8. Urinalysis (once pretest and in weeks 2, 5, & 13 in 10/sex/group)

Urine pH, volume, sodium, nitrogen, and potassium were increased and specific gravity was decreased in treatment groups (primarily MD and HD).

9. Organ weights

a) Kidney weights were dose-dependently increased (absolute weights +10 & +25% in HD males & females, respectively, compared to C).
b) Liver weights were D-D increased (absolute weights +25 & 60% in HD M & F, compared to C).

10. Gross Pathology

Gross findings included enlarged livers in 2 HD males, pale liver foci in 1 HD male, and liver masses in 1 HD female.

11. Histopathology (all tissues examined in C & HD males and females; liver and bladder only in LD & MD males; liver, bladder, and kidneys only in LD & MD females)

a) Liver: Centrilobular hepatocyte enlargement was noted in MD (7/15) & HD (15/15) males and MD (6/15) & HD (15/15) females.
b) Kidney: Renal lesions, consisting of transitional epithelial hyperplasia, microcalculus formation, and suppurative pyelonephritis, were increased in MD and HD females.
c) Urinary bladder: Hyperplasia of the transitional epithelium and suppurative cystitis were seen in HD females.

12. Drug exposure

Serum drug concentrations were determined in samples collected from 10/sex/group 24 hr after dosing during week 13. Because of the sample collection time and various analytical problems, the data are of limited value. Data indicate that trough levels were higher in females than in males. Measurable concentrations were found in less than 1/2 of males in each group, while most females had detectable levels. Mean trough concentrations were 23, 35, and 3.5X higher in females than in males at the LD, MD, and HD, respectively. In females, MD group plasma levels were higher than those measured in the HD group.

E) 3-MONTH ORAL TOXICITY IN RATS WITH 1-MONTH RECOVERY
(243819:1, GLP; Vol. 22)

1. Treatment

16/sex/group were dosed with 0, 10, 90, or 750 mg/kg/day, by gavage, for 3 months. At the end of treatment (day 91), 6/sex/group were retained until day 119 for evaluation of recovery.

Strain: Crj:CD(SD)(SPF)
Drug lot #: 9006522

2. Mortality

One HD male died on day 85; death was attributed to dosing error.

3. Clinical signs

Ataxia, amyotonia, lacrimation, soiling of lower abdomen and nose, and loss of righting were observed in HD animals.

4. Body weight and food consumption

a) BW gain was decreased in LD males (12%) and in HD males (15%) and females (20%) compared to C. Mean BWs at end of treatment were about 10% below C in these groups. No BW differences were seen after the recovery period.
b) Food consumption was decreased in the same 3 treatment groups. Water consumption was increased in MD males and in HD males and females. Increased water consumption remained in these groups after recovery.

5. Ophthalmoscopic examination

No treatment effects.

6. Hematology (10/sex on day 91 and 6/sex on day 119)

D-R decrease in prothrombin times seen in females, statistically significant at HD (-7% compared to C). No differences in recovery groups.

7. Clinical Chemistry (days 91 and 119)

a) Potassium decreased (10-20%) in HD groups.
b) Total protein and albumin slightly increased (10% compared to C) at HD.
c) Cholesterol (M 30%, F 130%), and phospholipids (M 20%, F 80%) increased at HD. Triglycerides decreased in HD males (40%) and increased in HD females (30%).
d) T4 decreased (10-25%) and TSH increased (15-60%) at MD & HD. (Possibly related to CA inhibition, ie, acetazolamide decreases uptake of iodine by thyroid.)
e) BUN increased (30-50%) at HD.
f) Alkaline phosphatase decreased (20%) in HD groups.
g) Bilirubin increased (30%) in HD males.
h) All values normal after 1 month recovery period.

8. Urinalysis (days 28, 91 and 119)

Urine pH and volume increased and specific gravity decreased in treatment groups (primarily at MD and HD). Changes not evident in recovery animals.

9. Organ weights

a) Liver weights were increased at the HD (absolute wts 15 & 50% above C in HD M & F, respectively) and remained elevated in recovery HD females.
b) Kidney weights were increased in HD animals (relative wts +25-30%) but were normal after recovery.
c) Thyroid weights were increased at the HD (rel wts +30-40%). No differences seen in recovery groups. (See clinical chemistry d), above).

10) Gross Pathology

No changes attributed to drug treatment.

11) Histopathology (all tissues examined in C & HD males and females at 3 months; liver, kidneys, stomach, Harderian gland, and bladder only in LD & MD at 3 months; liver, bladder, and kidneys only in recovery groups)

a) Liver: Centrilobular hepatocellular hypertrophy was noted in MD and HD groups. No liver changes seen in recovery groups.

b) Kidney: Hyperplasia of the transitional epithelium of the renal pelvis (very slight to moderate) was seen at the HD. Pyelonephritis and dilation of renal pelvis seen at MD and HD. Regeneration of renal tubule epithelium was observed in 1 LD and 1 HD female. Inflammatory cell infiltration was found in all treatment groups, but no dose-response was evident. No kidney changes were observed after recovery.

c) Urinary bladder: Hyperplasia of the transitional epithelium of the bladder mucosa (very slight to slight) was seen at MD and HD. Inflammatory cell infiltration of mucosa was increased in MD and HD animals. These changes remained in similar incidence and degree after the recovery period.

F) 12-MONTH ORAL TOXICITY IN RATS (A50771; GLP; Vol. 24)

1. Treatment

25/sex/group were dosed with 0, 10, 55, or 300 mg/kg/day, in the diet, for 12 months.
Strain: Wistar [Crl:COBS(WI)BR]
Drug lot #s: 8607560 & 8706545

2. Mortality

There were three HD deaths, but none considered treatment-related. One male was found dead during the 10th month (lymphosarcoma) and two females were sacrificed moribund in the 4th (ulcerated subcutaneous tumor) and 9th months (complications of orbital sinus puncture).

3. Clinical signs

No treatment-related clinical signs were observed.

4. Body weight and food consumption

a) Mean BW and BW gain were decreased in LD females (8 and 14%, respectively), MD males (7 and 10%) and females (12 and 21%), and HD males (11 and 16%) and females (17 and 30%).

b) Food consumption was generally decreased in treatment group rats, but the reduction was not considered of sufficient magnitude to account fully for the BW effects. There was a dose-dependent decrease in food efficiency (BW change divided by food consumption) throughout the study. During the last third of the study, mean food efficiency was decreased by 50% in the HD group; this was attributed to the antisecretory activity of topiramate and to the histopathological changes in the stomachs of treated rats.

5. Physical observations

There were no treatment-related morphological observations.

6. Ophthalmoscopic findings

There were no drug-related effects on the eye.

7. Hematology (weeks -2, 5, 13, 26, 40 & 52)

Small, dose-dependent decreases (maximal 10-15% below C) in RBCs, HGB, and HCT were seen throughout the study in treatment group rats compared to C. Erythrocyte indices were not consistently altered, and no other hematological changes were observed.

8. Clinical Chemistry (weeks -2, 5, 13, 26, 40 & 52)

a) Increased (3-5%) serum chloride values were observed in all topiramate-treated groups throughout study.
b) Cholesterol was increased (40%) in HD females, and triglyceride values were decreased (35-40%) in HD males and females.
c) ALT and AST were decreased (40-50%) in MD and HD females.

9. Urinalysis

Not performed.

10. Organ weights

a) Increases in absolute (16%) and relative (42%) liver weights were seen in HD females.
b) Absolute and relative kidney weights were increased in HD males (13 & 27%, respectively) and MD (7 & 21%) and HD females (24 & 54%).

11. Morphological Pathology (routine histopathology in C & HD; gross lesions, liver, stomach, kidney, and urinary bladder in other groups)

Treatment-related changes in the stomach, kidneys, ureters, urinary bladder, and liver were observed.

a) Stomach - Changes in the fundic mucosa were seen in all treatment groups, their incidence and severity increasing with dose. These consisted of hyperplasia of the generative cell zone of the neck of the gastric gland (**Table III.1**), and associated reductions in the parietal cell mass (frank degeneration at HD) and increases in the foveolar epithelium. The incidence and severity of dilated deep fundic glands was increased slightly in HD females. These changes were accompanied by inflammatory cell infiltration. The proliferative changes were not considered preneoplastic by the sponsor or by a consulting pathologist.
b) Urinary tract - D-R increases in the incidence and severity of phosphate urolithiasis, urothelial hyperplasia (**Table III.2**), papillary collecting duct epithelial hypertrophy with cytoplasmic acidophilic droplets, and focal mineralization (nephrocalcinosis) involving proximal convoluted tubules (in males) were seen in treated rats, primarily at MD & HD. Urothelial hyperplasia was considered to be a secondary reaction to renal calculi or mineralized mucosa acting as foreign bodies (**Table III.3**). Calculi were seen grossly in 3 HD males and in all female groups, with an increased incidence in MD & HD. The calculi were composed of calcium

and magnesium phosphate with protein matrices and did not contain topiramate. None of the hyperplasias were thought to represent preneoplastic lesions.

c) Liver - The incidence of centrilobular hepatocytic hypertrophy was increased in MD & HD males and females (0, 1, 7, & 14/25 in C, LD, MD, & HD males, respectively; 1, 1, 5, & 16/25 in C, LD, MD, & HD females, respectively).

d) Tumors - No treatment-related effects. HD tumors not considered treatment-related included myxomatous mesenchymal tumor in female sacrificed on day 130, lymphosarcoma in male found dead on day 309, and renal cell adenoma in 1 male (rare tumor in rats).

Table III.1: Incidence and Severity of Gastric Generative Zone Hyperplasia

	Male				Female			
Group (mg/kg)	0	10	55	300	0	10	55	300
Number Examined	25	25	25	25	25	25	25	25
Total Hyperplasia	3	12**	19**	24**	0	3	10**	25**
Focal Hyperplasia	3	10*	6	0	0	3	3	2
Minimal	2	4	0	-	-	0	1	0
Mild	1	6	6	-	-	3	2	2
Diffuse Hyperplasia	0	2	13**	24**	0	0	7**	23**
Mild	-	2	11	2	-	-	6	2
Moderate	-	0	2	21	-	-	1	18
Marked	-	0	0	1	-	-	0	3

*p<0.05, **P<.001

Table III.2: Incidence and Severity of Urothelial Hyperplasia

	Male				Female			
Group (mg/kg)	0	10	55	300	0	10	55	300
Kidneys Examined	25	25	25	25	25	25	25	25
Renal Urothelial Hyperplasia	6	2	6	16**	10	6	17*	17*
Minimal	5	2	4	7	9	3	8	5
Mild	1	0	2	7	1	3	5	8
Moderate	0	0	0	2	0	0	4	4
Ureters Examined	0	1	1	0	0	0	0	2
Ureter Urothelial Hyperplasia	-	0	0	-	-	-	-	2
Mild	-	-	-	-	-	-	-	1
Moderate	-	-	-	-	-	-	-	1
Bladders Examined	25	25	25	25	25	25	25	23
Bladder Urothelial Hyperplasia	0	0	2	3	0	0	1	2
Minimal	-	-	0	0	-	-	1	0
Mild	-	-	2	2	-	-	0	1
Marked	-	-	0	1	-	-	0	1

*p<0.05, **P<.001

Table III.3: Association between Renal Urothelial Hyperplasia and Renal Calculi

	Male				Female			
Group (mg/kg)	0	10	55	300	0	10	55	300
Number Examined	25	25	25	25	25	25	25	25
Urothelial Hyperplasia	6	2	6	16	10	6	17*	17*
Gross Calculi	0	0	0	2	1	2	7*	8*
Associated Hyperplasia	-	-	-	2	1	2	7*	7*
Microcalculi	1	1	5	2	13	11	18	15
Associated Hyperplasia	0	0	4	2	7	6	16*	13
Mineralization	1	1	5	12**	11	3*	9	5
Associated Hyperplasia	1	1	4	10**	7	1*	6	3
Non-associated Hyperplasia	5	1	1	4	0	0	0	2

*p<0.05, **P<.001

12. Drug exposure

In a satellite toxicokinetics study (A500926, Vol. 42), Wistar rats (5/sex/group) received topiramate doses of 10, 55, or 300 mg/kg/day in the diet for 1 year. Single blood samples were collected 1-2 hr into the light cycle at 0.25, 6, 9, and 12 months. At 3 months, samples were collected over 24 hr for AUC determinations:

Dose (mg/kg/day)	AUC (ug·hr/ml)	
	Male	Female
10	27.8 ± 8.8	79.3 ± 7.3
55	144.9 ± 37	361.4 ± 27.9
300	356.4 ± 43.4	1083 ± 256.1

Levels present during 24-hr sampling at 3 months indicate nearly continuous exposure at all doses, but with considerable circadian variation. Female AUC values were greater (2.5-3-fold) than those for males at each dose level. An approximately proportional increase in exposure occurred between the LD and MD in males, but the increase was less than proportional between the MD and HD. In females, exposure increases were less than dose-proportional between all doses. In serum levels measured at 1-2 hr into the light cycle over the course of the study, there were no obvious changes between 1 week and 12 months.

G) 11-MONTH RAT EXPLORATORY AND REVERSIBILITY STUDY (243792:1; Vol. 25)

1. Treatment

26 male and 23 female Wistar rats were treated (in diet) with >300 (320-750) and >450 (450-1100) mg/kg/day, respectively, for approximately 11 months. Untreated controls (20M, 21F) were included.

2. Results

At 11 months, mean BWs were decreased by 21 and 29% in treated males and females, respectively, and serum gastrin levels were about 2-fold higher than in C. Pathology findings in the stomach were essentially the same as in the previous 1-year study: increased mucous over fundic mucosa, hyperplasia of the generative zone at neck of the gastric mucosal gland, reduction in parietal cell mass, increase in foveolar epithelium, and inflammatory cell infiltration. Reversal of gastrin elevation and histomorphological alterations was seen at 9 and 20 weeks after treatment withdrawal. At no time were enterochromaffin-like cells increased.

H) 3-MONTH ORAL TOXICITY IN DOGS (A47180; GLP; Vol. 25)

1. Treatment

4/sex at 0, 10, 40, or 150 mg/kg/day, in gelatine capsules.
Drug lot #: 8420100

2. Mortality

No deaths.

3. Clinical signs

No treatment effects.

4 Body weight and food consumption

Mean BW gain decreased in MD and HD males (10% less than C), and HD females lost 7% compared to 12% gain in C. Food consumption and food efficiency decreased in MD males and HD males and females.

5. Physical examinations (once pre-test and weeks 13)

There were no treatment-related observations.

6. Ophthalmoscopic examinations (pre-test and week 13)

There were no drug-related effects on the eye.

7. Cardiovascular examinations (twice pre-test and weeks 13 & 14)

No drug effects.

8. Hematology (weeks -4, -2, 3, 5, 8, & 13)

Dose-dependent decreases in RBCs, HCT, and HGB were seen throughout treatment (10-15% at HD). RBC indices and platelets were somewhat increased at HD compared to controls. There was no morphological evidence of extramedullary hematopoiesis, increased hemosiderin deposition, or bone marrow depletion.

9. Clinical Chemistry (weeks -4, -2, 3, 5, 8, & 13)

a) BUN increased in treated animals compared to C and baseline (15-20% at HD). (Increased BUN may be associated with metabolic acidosis during diuresis.)
b) Alkaline phosphatase increased at HD (2-2.5X C), but no morphological correlates.
c) Total protein, albumin, and A/G ratios decreased in treated animals.
d) Serum potassium decreased (10% below C at HD) and chloride increased (5% at HD) dose-dependently.
e) Transaminase levels decreased in treatment groups (SGPT & SGOT were 35 & 50% below C, respectively, in HD males).

10. Urinalysis (weeks -4, -2, 3, 5, 8, & 13)

a) Urine pH increased in all treatment groups compared to controls.
b) Specific gravity slightly decreased in treatment groups.

11. Organ weights

Increases in absolute and relative liver weights in MD females and in HD males and females.

12. Morphological Pathology (complete histopath on C & HD; stomach & bladder in all groups)

No treatment-related lesions.

13. Drug Exposure

Serum concentrations of topiramate were determined in samples (4/sex/group) obtained 24 hr after dosing during weeks 3 and 13 and 1 hr after dosing during week 14. 24 hr levels were at or below the quantification limit (0.25 ug/ml) in most cases. Levels measured at about 1 hr after dosing during week 14 were consistent with PK data in dogs; levels increased with dose (non-proportional) and were similar between sexes at each dose (HD data limited, however):

Dose (mg/kg/day)	Mean Concentration (ug/ml)	
	Male	Female
10	8.8 ± 0.2	7.8 ± 1.3
40	17.8 ± 7.4	19.1 ± 3.6
150	105.5 ± 23.7	130.2 (only 1 sample)

I) 12-MONTH ORAL TOXICITY IN DOGS (A50656; GLP; Vol. 25)

1. Treatment

4/sex/group were dosed with 0, 10, 30, or 100 mg/kg/day, in gelatine capsules.
Drug lot #: 8607560

2. Mortality

No deaths.

3. Clinical signs

Emesis observed at all dose levels.

4. Body weight and food consumption

a) Mean BW gain in HD males and females was 13 and 20%, respectively, below controls.
b) Food consumption was decreased in HD females.

5. Physical examinations (once pretest and weeks 26 & 52)

There were no treatment-related observations.

6. Ophthalmoscopic examinations (pretest and weeks 26, 40, & 53)

There were no drug-related effects on the eye.

7. Cardiovascular examinations (weeks -7/-6, -2/-1, 26, & 52)

No drug effects.

8. Hematology (weeks -7/-6, -5, -2, 5, 13, 26, 40 & 53)

Small D-R decreases in RBC, HCT, and HGB were seen throughout treatment (10-20% at HD). One HD female (3234) had more severely depressed HCT, HGB, MCH, & MCV values with increased platelets. Generally, RBC indices and platelets were somewhat increased compared to controls.

9. Clinical Chemistry (weeks -7/-6, -5, -2, 5, 13, 26, 40 & 53)

a) Alkaline phosphatase increased (up to 2X C at HD).
b) Small decreases in total protein, albumin, and A/G ratios.
c) Serum potassium dose-dependently decreased (HD 10-15% below baseline at 53 weeks) and chloride increased (HD 5-10% above baseline at 53 weeks).
d) Cholesterol generally increased (up to 40% above C at HD).
e) Small dose-dependent decreases in transaminase levels generally seen; however, SGPT increased to 16X C or baseline in 1 HD male (3230) at 53 weeks.

10. Urinalysis (weeks -7/-6, -5, -2, 5, 13, 26, 40 & 53)

Urine pH was increased in all treatment groups compared to controls. Urine volume was increased at the HD. These effects were attributed to the pharmacological action of drug, ie, inhibition of carbonic anhydrase. There was a possible increase in hematuria among treated females, and crystalluria appeared slightly increased in treated dogs.

11. Organ weights (liver, kidneys, spleen, heart, and brain weighed)

a) Increase in absolute (10-15% above C at HD) and relative (30% at HD) liver weights in treatment groups.
b) Decrease in absolute kidney weights (10% below C at HD).
c) Decrease in absolute (20-40% below C at HD) and relative spleen weights (15-30% below C at HD).

12. Morphological Pathology (routine histopathology on C & HD only; gross lesions, ovaries, uteri, & stomach examined in other groups)

No gross of microscopic lesions considered treatment-related by the sponsor were noted. However, focal mineralization of the kidney was seen in 3/4 HD females vs 1/4 C females.

IV. CARCINOGENICITY

A) TWO YEAR CARCINOGENICITY STUDY IN MICE (A500736; GLP; Vols. 29-33)

1. Treatment

Mice were dosed with 0 (C I), 0 (C II), 20, 75, or 300 mg/kg, in the diet, for 53 weeks (10/sex/group) or 93-95 weeks (60/sex/group). The HD was based on the MTD in a 6-month pilot study; the LD is the estimated daily therapeutic dose. Study duration was shortened to ensure adequate numbers for evaluation.

Strain: Crl: CD-1(ICR)BR, VAF/Plus
Drug Lot #: 8806672

2. Mortality

Study mortality rates were approximately 50-70%. No treatment effect was observed in males. Survival was decreased in HD females compared to C I, but not compared to C II or combined controls. (See statistical review).

Mortality Incidence

Dose (mg/kg)	Male (Week 94)	Female (Week 93)
0 (control I)	36	31
0 (control II)	35	42
20	36	36
75	29	38
300	32	44

3. Observed Signs

No treatment-related clinical signs. Staph infection was found in all groups.

4. Body Weight

BW means were intermittently lower (5-10%) in treatment groups compared to controls throughout the study; however, there were no differences in terminal BWs.

5. Food Consumption

No drug-related changes in food consumption.

6. Tissue Mass Observations

No drug-related changes in palpable masses.

7. Clinical Pathology

Only serum gastrin was measured, at the interim and terminal sacrifices. Mean gastrin levels were increased (40%) in HD females at study termination.

8. Gross Pathology

a) 1-Year Interim Sacrifice

No gross findings related to treatment.

b) Terminal Sacrifice

i. Stomach - Thickened mucosa was found in 1/60 MD and 3/60 HD males. A nodule was observed in 1 HD male.
ii. Kidney - Incidences of renal discoloration, enlargement, capsular irregularities, and pelvic dilatation or hydronephrosis were increased in treated females. Uroliths were seen in one MD and one HD female.
iii. Urinary bladder - Distension was observed only in treated females, in 4/60, 5/60, and 9/60 of LD, MD, and HD mice, respectively.
iv. Spleen - The incidence of splenomegaly was increased in treated males.

9. Microscopic Pathology

Complete microscopic examinations were performed on all groups except C I. Examination of C I was limited to the urinary bladder.

a) 1-Year Interim Sacrifice

i. Liver - Hepatocellular hypertrophy was seen in 1/59, 2/60, 5/60, and 8/60 males in the C II, LD, MD, and HD groups, respectively.
ii. Stomach (body) - The incidence of dilated gastric glands was increased in treated males and females. Incidences of lymphocytic infiltration and amyloidosis of the gastric mucosa were increased in treated females. A hyperplastic diverticulum was found in the submucosa in one HD male.
iii. Kidney - The incidence of tubular dilatation was increased in HD females.
iv. Urinary bladder - No proliferative changes were found at 1 year.

b) Terminal Sacrifice

Non-neoplastic

i. Liver - The incidence of hepatocellular hypertrophy was increased in HD males (27/60) and MD (5/60) and HD females (18/60) compared to controls (9/59 males, 0/60 females). The incidence of hepatocellular hyperplasia was increased in HD males (6/60 vs 1/59 in C). Hepatic extramedullary hematopoiesis was increased in treated females.
ii. Stomach (body) - The incidence of gastric mucosal (generative-cell zone) hyperplasia was increased in MD and HD males (33/60 & 45/60) and females (20/60 & 38/60) compared to C (1/59 & 2/60 in M & F). The incidence of focal hyperplastic diverticula was increased in HD males (9/60 vs 3/59 in C).
iii. Kidney - Incidences of renal pelvic dilatation and chronic pyelonephritis or chronic glomerulonephritis were increased in HD females.
iv. Urinary bladder - Increased incidences of chronic or hyperplastic cystitis (5/59 HD♀s vs 0/120 C), focal mucosal hyperplasia (6/60 HD♂s vs 0/117 C; 4/60 MD, 6/59 HD♀s vs 1/120 C), calculi (2/59 HD♀s vs 0/120 C), and

dilatation of the lumen (2/57, 3/60, & 5/59 in LD, MD, & HD♀s, respectively, vs 0/120 C) were observed in treated mice.

v. Spleen - The incidence of splenic hyperplasia was slightly increased in the HD group.

Neoplastic

i. Urinary bladder - Tumor incidence was increased in HD males and in females from all treatment groups, largely due to the increased occurrence of leiomyosarcomas (**Table IV.1**). Statistical significance was reached for leiomyosarcomas in HD males and females, and for all bladder tumors in HD females. In the FDA statistical analysis, there were significant survival adjusted positive linear trends in the tumor incidence rates of leiomyosarcoma in both males and females.

ii. Other - Increased (but low) incidences of the following tumors were observed in drug-treated groups: squamous cell carcinoma of (fore)stomach in males (0, 1, 1, & 2 in C II, LD, MD, & HD, respectively), uterine hemangioma (1, 0, 2, & 3 in C II, LD, MD, & HD), uterine leiomyosarcoma (2 HD), cervical leiomyoma (3 MD, 2 HD), lung bronchiolo-alveolar carcinoma in females (0, 2, 1, 2 in C II, LD, MD, & HD), ovarian hemangioma (1 HD), splenic hemangiosarcoma (1 LD male, 1 MD & 1 HD female), malignant meningioma (1 HD male), hepatocellular carcinoma in females (1 HD), and renal tubular carcinoma (1 HD male). Some of these were statistically significant in the sponsors analysis but not in the FDA analysis.

Table IV.1: Incidence of Primary Neoplasms in the Mouse Urinary Bladder

Dose (mg/kg/day)	0	0	20	75	300	0	0	20	75	300
Sex:	M	M	M	M	M	F	F	F	F	F
No./Group:	59	59	60	60	60	60	60	60	60	60
Tumor Type:										
carcinoma (transitional cell)	0	0	0	0	0	0	0	0	0	1
hemangiosarcoma	0	0	0	0	0	0	0	0	0	1
leiomyoma	0	0	0	0	0	0	1	0	0	0
leiomyosarcoma	0	1	0	0	4	1	0	3	1	6
papilloma	0	0	0	0	0	0	0	0	0	1
polyp, stromal	0	0	0	0	0	0	0	0	1	0
Total Incidence	0	1	0	0	4	1	1	3	2	9

10. Drug exposure

In a separate toxicokinetics study (239843:1, Vol. 42), CD-1 mice (27/sex/group) received topiramate doses of 20, 75, or 300 mg/kg/day, in the diet, for 1 month. Terminal blood samples were collected from 3/sex/group every 3 hr for 24 hr, and plasma drug levels were measured.

Data indicated nearly continuous exposure at all doses, although levels showed considerable circadian variation. Male plasma AUC values were greater (1.7-4.5-fold) than those for females at each dose level. Approximately proportional increases in female AUCs were seen between the LD and MD, but the increase was much greater than proportional between the MD and HD. In males, exposure increased less than dose-proportionally between the LD and HD, but proportionally between the MD and HD.

Dose (mg/kg/day)	Plasma AUC (ug·hr/ml; mean ± SEM)	
	Male	Female
20	11.2 ± 0.7	2.5 ± 0.4
75	55.1 ± 4.7	12.6 ± 1.8
300	225.3 ± 15.7	133.2 ± 14.7

B) TWO YEAR CARCINOGENICITY STUDY IN RATS (A500737; GLP; Vols. 26-28)

1. Treatment

50 rats/sex were dosed with 0 (C I), 0 (C II), 20, 45, or 120 mg/kg, in the diet, for 104-105 weeks. The HD was based on toxicity in the 12-month oral toxicity study; LD is the estimated daily clinical dose.

Strain: Wistar [Crl:(WI)BR, VAF/Plus]
Drug Lot #: 8806672

2. Mortality

Survival was not increased by drug administration (see FDA statistical review).

Mortality Incidence

Dose (mg/kg)	Male	Female
0 (contol I)	20	24
0 (control II)	28	22
20	19	18
45	15	17
120	15	22

3. Observed Signs

No drug-related clinical signs noted.

4. Body Weight

Decreased in MD and HD females (final means 6 & 21%, respectively, below C II) and in HD males (7% below C II). Smaller decreases seen in LD females, primarily between weeks 23 and 81, and in LD & MD males, primarily during the first year.

5. Food Consumption

No drug-related effects in males. Decreased in HD females throughout most of study.

6. Ophthalmoscopic Exam

No treatment-related effects.

7. Tissue Mass Observations

No drug-related changes in number of palpable masses.

8. Clinical Pathology

Only gastrin levels were measured. Mean serum gastrin levels were increased compared to control (C I) levels in HD (70%) males at 57 weeks and in MD (40%) and HD (100%) males and HD females (50%) at week 105.

9. Gross Pathology

Pleural foci in the lungs were increased in HD males. Increased numbers of HD males and females had mucoid stomach contents. A prominent limiting ridge was noted in the stomachs of several HD males. The incidence of renal pelvic calculi was increased in males and females from all treatment groups. Sediment in the urinary bladder lumen and dilatation of the bladder were increased in HD males.

10. Microscopic Pathology

Complete microscopic examinations were performed on all groups except C I.

a) Non-neoplastic

i. Liver - Increased incidences of centrilobular hepatocellular hypertrophy (0, 3, 16, 16/50 in C, LD, MD, HD males, respectively; 0, 0, 17, 33/50 in corresponding females), vacuolization (9/50 MD, 7/50 HD vs 2/50 C females), and eosinophilic foci (23/50 HD vs 10/50 C males; 5, 15, 16, 15/50 in C, LD, MD, HD females, respectively) were found in treated rats.

ii. Stomach - Increased incidences of hyperplasia of the generative cell zone of the fundic gastric glands (5, 6, 24, 45/50 in C, LD, MD, HD male, respectively; 4, 8, 39, 47/50 in corresponding females), decreased parietal cells, inflammatory cell infiltration, and hyperplasia of the limiting ridge between the forestomach and fundus were found in MD and HD groups.

iii. Kidney - The incidence of renal calculi was increased in HD males (23/50 vs 10/50 in C) and in treated females (38, 43, 50, 47/50 of C, LD, MD, HD, respectively). D-R increase in severity in both sexes. Incidence of urothelial hyperplasia was increased in HD males (23/50 vs 13/50 C) and

in females at all doses (24, 31, 35, 29/50 in C, LD, MD, HD, respectively).

iv. Urinary bladder - Incidences of calculi (2/50 HD M, 1/50 HD F), mucosal hyperplasia (0/50, 1/49, 2/49, 3/50 in C, LD, MD, HD males, respectively; 2, 1, 1, 4/50 in respective females), and dilation of the urinary bladder (4, 3, 7, 8 in C, LD, MD, HD males, respectively) were higher in treated rats.

b) Neoplastic

There were no statistically significant increases in tumors (see statistical review).

i. Urinary bladder - Mucosal papillomas were observed in one male in each of the MD and HD groups and in one HD female, possibly secondary to chronic irritation induced by calculi in the lumen. A leiomyosarcoma was found in 1 MD female.

ii. Other - Single incidences of renal tubular carcinoma (1 HD female) and squamous cell carcinoma of (fore)stomach (1 HD female) were found only in treatment groups.

V. GENETIC TOXICOLOGY (Vol. 38)

A) AMES TEST (A44203; GLP)

When topiramate (5-5000 ug/plate) was tested in strains TA98, TA100, TA1535, TA1537, and TA1538, directly and with metabolic activation, no increases in revertant colonies were observed.

B) REVERSE MUTATION IN BACTERIA (243688:1, conducted by Kyowa; GLP)

When topiramate (156-5000 ug/plate) was tested in a pre-incubation assay using Salmonella TA98, TA100, TA1535, TA 1537, and E. coli WP2uvrA, in the absence and presence of S-9, no increases in revertant colonies were seen.

C) MOUSE LYMPHOMA ASSAY (A44401; GLP)

When topiramate (1-5000 ug/ml) was tested in a L5178Y/TK+/- mouse lymphoma cell forward mutation assay, with and without S-9, no increases in mutation frequency were observed.

D) RAT HEPATOCYTE DNA REPAIR ASSAY (A44655; GLP)

No increases in unscheduled DNA synthesis (autoradiographic quantification) in primary cultures of adult rat hepatocytes at topiramate concentrations of 100, 500, 1000, 2000, and 2500 ug/ml (-.045, -0.62, -0.68, -0.81, -0.59 mean net nuclear grains, respectively). Positive (1 ug/ml 2-acetylaminofluorene; 12.65 mean net nuclear grains) and negative (1% DMSO; -0.29 mean net nuclear grains) controls were adequate.

E) HUMAN LYMPHOCYTE ASSAY (A500004, conducted by Microbiological Associates; GLP)

Human peripheral lymphocytes were exposed in culture for 24 hr without S-9 or 4 hr with S-9 to topiramate concentrations of 63, 125, 250, and 500 ug/ml (without S-9) or 625, 1250, 2500, and 5000 ug/ml (with S-9). Twenty four hours after initiation of treatment, cell division was arrested and metaphase chromosome spreads (100/treatment) were examined for aberrations. No increases in chromosome aberrations were observed. Mitotic index was reduced by 62 and 57%, relative to solvent (DMSO), at 500 ug/ml (minus S-9) and 5000 ug/ml (plus S-9), respectively. Positive controls (mitomycin C and cyclophosphamide) gave adequate responses.

F) IN VIVO BONE MARROW ASSAY IN RATS (A46698; GLP)

Wistar rats (5/sex/time/group) were given doses of 100, 400, or 1000 mg/kg po of topiramate (HD about 1/4 male LD50 and 1/3 female LD50), and bone marrow was sampled 6, 24, or 48 hr after exposure. Groups given triethlenemelamine (5/sex) and vehicle (10/sex) served as positive and negative controls, respectively. After induction of metaphase accumulation with colchicine, femoral bone marrow preparations from the HD topiramate group and positive and negative control groups were evaluated for alterations of chromosome structure and number. Results are summarized in **Table V.1**.

Dose-related increases in clinical signs were observed in topiramate-treated groups. Mitotic index values were decreased in treatment groups (all times) and in positive control groups, indicating bone marrow toxicity. Small increases (not statistically significant) in frequency of chromosome aberrations (12/360 vs 4/600) and in aberrant cells (10/360 vs 3/600) compared to controls were observed in male treatment groups at 6 hr, due primarily to one animal with 10 aberrations (8

chromatid breaks, 2 chromosome breaks), 8 aberrant cells, and 31 gaps in 120 analyzed cells. A slight increase (not statistically significant) in aberrations was also seen in female treatment group animals at same time after treatment (3/360 vs 1/600 in C). Results were considered negative. Positive controls showed appropriate responses.

Table V.1: Summary Data from in vivo Cytogenetic Evaluation of Topiramate in Wistar Rats

MALES

Group	T[1]	No. of Rats/ Group	Mitotic Index per 1000 Nucleated Cells Mean (± S. E.)	Non-diploid Cells / Cells Scored	Chromosomal Aberrations[2] / Cells Scored	Aberrant Cells[2] / Cells Scored
Negative Control (p.o.)						
0 mg/kg McN-4853	24	10	37.3 (± 3.8)	3/600	4/600	3/600
Single Acute Dose (p.o.)						
1000 mg/kg McN-4853	6	5	44.7 (± 10.2)[3]	0/360	12/360	10/360
1000 mg/kg McN-4853	24	5	49.9 (± 11.2)	6/336	0/336	0/336
1000 mg/kg McN-4853	48	5	46.8 (± 3.5)[3]	4/300	0/300	0/300
Positive Indicator (i.p.)						
0.3 mg/kg TEM	24	5	43.0 (± 10.5)[3]	6/319	483/360	86/360

FEMALES

Group	T[1]	No. of Rats/ Group	Mitotic Index per 1000 Nucleated Cells Mean (± S. E.)	Non-diploid Cells / Cells Scored	Chromosomal Aberrations[2] / Cells Scored	Aberrant Cells[2] / Cells Scored
Negative Control (p.o.)						
0 mg/kg McN-4853	24	10	50.7 (± 4.9)	6/600	1/600	1/600
Single Acute Dose (p.o.)						
1000 mg/kg McN-4853	6	5	37.8 (± 8.0)[3]	4/360	3/360	3/360
1000 mg/kg McN-4853	24	5	41.6 (± 3.1)	2/300	1/300	1/300
1000 mg/kg McN-4853	48	5	47.8 (± 6.2)	1/300	1/300	1/300
Positive Indicator (i.p.)						
0.3 mg/kg TEM	24	5	26.3 (± 8.6)[4]	4/305	151/315	52/315

1 T: approximate interval (in hours) between dosing and sacrifice (see Methods).
2 Includes chromosome aberrations, but not gaps or numerical deviations.
3 $p \leq 0.05$, Student's t-Test, one-tailed, as compared with negative control values.
4 $p \leq 0.01$, Student's t-Test, one-tailed, as compared with negative control values.

VI. REPRODUCTIVE TOXICOLOGY

A) SEGMENT I STUDY IN MALE RATS (A50270; GLP; Vol 34)

1. Treatment

Male rats (26/grp) were dosed with 0, 0.2, 8, 25, or 100 mg/kg, by gavage, for 70 days prior to mating and for 30 days after initiation of mating (1:1 cohabitation with untreated females), then sacrificed. Females were sacrificed either on day 13 of gestation (1/2) or day 21 of lactation.

Strain: Sprague-Dawley [Crl:COBS CD(SD)BR]

2. Fo Data

a) No deaths due to drug-treatment.
b) No drug-related clinical signs noted.
c) Small dose-dependent decreases in male BW, BW gain, and food consumption were observed. Weight gain over the entire treatment period was about 10% below C at 25 and 100 mg/kg.
d) There was a trend toward increasing absolute and relative testes weights with increasing dose in F0 males at necropsy; this was attributed to random variation and the BW effect, but no histopathology was performed.
e) Small (not statistically significant) increases in days of cohabitation to mating were observed in treated males. (Decreased libido has been reported with therapeutic use of acetazolamide).
f) There were no treatment-related effects on fertility (pregnancy index, mating index, or fecundity index; **Table VI.1**).

Table VI.1: Fertility Data

	Sire's Dosage of Topiramate (mg/kg)				
	0	0.2	8.0	25	100
No. of Females	25	26	25	24	26
Pregnancy Index[1]	22/27 (81%)	23/26 (88%)	25/27 (93%)	24/28 (86%)	25/26 (96%)
Mating Index[2]	26/27 (96%)	26/26 (100%)	27/27 (100%)	27/28 (96%)	26/26 (100%)
Fecundity Index[3]	22/26 (85%)	23/26 (88%)	25/27 (93%)	24/27 (89%)	25/26 (96%)

[1] Pregnancy Index = $\frac{\text{Number Pregnant}}{\text{Total Cohabited}} \times 100$

[2] Mating Index = $\frac{\text{Number Copulated}}{\text{Total Cohabited}} \times 100$

[3] Fecundity Index = $\frac{\text{Number Pregnant}}{\text{Number Copulated}} \times 100$

3. Reproductive parameters

a) Small increases in pre- and postimplantation loss were seen in treatment group litters at day 13. There was no dose-response for preimplantation loss, but resorptions and postimplantation loss were increased in a dose-related manner. (HD mean resorption value (1.08) outside historical control range (0.53-0.88); however, no concomitant decrease in live embryos seen).
b) There were no treatment-related effects on reproductive parameters in dams allowed to deliver (gestation length, uterine implants on day 21 of lactation, viable pups), and no effects on the F1 generation.

B) SEGMENT I STUDY IN FEMALE RATS (A50602; GLP; Vol. 35)

1. Treatment

Female rats (29/grp) were dosed with 0, 0.2, 8, 25, or 100 mg/kg, by gavage, for 14 days prior to mating, during the mating period (1:1 cohabitation with untreated males for up to 19 days), and until termination on either day 13 of gestation (1/2 were sacrificed at this time) or Day 21 of lactation.

Strain: Sprague-Dawley [Crl:COBS CD(SD)BR]
Drug lot #: 8706545

2. Fo Data

a) No deaths due to drug-treatment.
b) No drug-related signs noted.
c) Weight gain and food consumption were decreased dose-dependently in treatment groups during the first week of dosing prior to mating. No differences thereafter.
d) Mating, pregnancy, and fecundity were not affected by drug treatment.
e) There was an increase in the observations of granular material in the renal pelvis/urinary bladder and of dilated renal pelvis in F0 females at necropsy.

3. C-Section Data **(Table VI.2)**

a) Corpora lutea and implants were decreased (significant at LD and HD). Control values were said to be higher than in historical controls, but the effect appears dose-dependent at all but LD.
b) Post-implantation loss was slightly increased at 25 and 100 mg/kg (7.3 & 7.6%, respectively, vs 4.3% in C).

4. Delivery Data

a) No effects on gestation length, implantation sites (determined on P21), litter size, or pup viability were seen at birth.
b) No drug-related increase in postnatal mortality was observed.
c) Pup weights were decreased slightly in treated litters at birth; this deficit persisted in HD litters (mean BW 8% below C on P21).
d) No gross abnormalities were observed in dead pups or pups at necropsy.

Table VI.2: Gestation Day 13 Caesarean Section Data (mean ± SE)

	Dose (mg/kg)				
	0	0.2	8.0	25	100
Number of Litters	11	14	12	14	12
Corpora Lutea	17.1 ± 0.6	13.7 ± 1.6**	16.5 ± 0.4	14.9 ± 1.0	14.3 ± 0.5**
Uterine Implants	16.6 ± 0.6	13.1 ± 1.1**	15.9 ± 0.4	14.4 ± 1.0	14.3 ± 0.4**
Embryos	15.8 ± 0.7	12.6 ± 1.1	14.8 ± 0.5	13.4 ± 1.0	13.7 ± 0.5
Resorption	0.7 ± 0.2	0.5 ± 0.2	1.2 ± 0.3	0.9 ± 0.2	0.6 ± 0.2
Pre-implantation Loss	3.2 ± 1.2	4.1 ± 2.1	3.5 ± 1.2	3.6 ± 1.5	0.5 ± 0.5
Post-implantation Loss	4.3 ± 1.6	4.5 ± 1.5	4.1 ± 2.2	7.3 ± 2.0	7.6 ± 2.1

** Significantly different from control, $p \leq 0.01$

C) TERATOLOGY STUDY IN MICE (A47164; GLP; Vol. 35)

1. Treatment

26 F/group were dosed with 0, 20, 100, or 500 mg/kg, by gavage, on days 6 through 15 of gestation. C-sections were performed on day 18 of gestation.

Strain: CD-1
Drug lot #: 8420100

2. Maternal Effects

No drug-related changes in mortality or clinical signs. Mean BW was slightly decreased on days 15 and 18 in the HD group. BW gain was decreased (20%) over days 6-15 and 6-18 at the HD.

3. C-Section Data

Small increases in intrauterine death were seen in MD and HD litters (1.5 and 1.3 mean resorptions/litter, respectively, vs 1.1 in C), including 1 total litter loss at the HD. Fetal weights were decreased (4 & 10%, respectively) and the number of small fetuses (<1 g) was increased at the MD and HD.

4. Fetal Evaluations

All fetuses examined for external malformations, 1/2 for visceral (Wilson's) and 1/2 for skeletal defects.

a) Major malformations were increased at all doses. Malformed fetuses were found in 8.3, 25, 35, and 32% of C, LD, MD, and HD litters, respectively, and numbers of malformed fetuses increased dose-dependently **(Table VI.3)**.

b) Craniofacial malformations were increased in all treated groups. Single incidences of exencephaly were seen at the MD and HD only. Limb reduction defects were found in a single fetus at the LD only (CD-1 is considered resistant to this effect).

c) The incidence of skeletal variations (primarily reduced ossification) was increased at the HD.

Table VI.3: Summary of Fetal Malformations in Mice

	Dose (mg/kg)			
	0	20	100	500
No. of litters examined	24	24	26	22
No. of fetuses examined externally	272	276	285	236
No. of fetuses examined viscerally	140	142	145	122
No. of fetuses examined skeletally	132	134	140	114
Malformations in fetuses (litters)				
Cleft palate	1 (1)	2 (2)	4 (3)	6 (2)
Limb and/or digit abnormalities	0	1 (1)	1 (1)	0
Interventricular septal defect	0	1 (1)	0	0
Reduced lower jaw	0	0	1 (1)	0
Microstomia	0	0	1 (1)	0
Exencephaly	0	0	1 (1)	1 (1)
Ablepharon	0	2 (2)	2 (2)	3 (3)
Thoracic hernia w/ sternal cleft	0	0	0	1 (1)
Partial situs inversus	0	0	1 (1)	0
Agenesis of one kidney	0	0	1 (1)	0
Folded retina	1 (1)	1 (1)	1 (1)	2 (1)
Hydronephrosis	1 (1)	0	0	1 (1)
Total fetuses (litters) with malformations	3 (2)	6 (6)	10 (9)	13 (7)

D) TERATOLOGY STUDY IN RATS (McNeil Study A47091; GLP; Vol. 35)

1. Treatment

52 pregnant females were dosed with vehicle and 26 females/group dosed with 20, 100, or 500 mg/kg, by gavage, on days 6 through 15 of gestation. C-sections were performed on day 20 of gestation.

Strain: Sprague-Dawley [Crl:COBS CD(SD)BR]
Drug lot #: 8420100

2. Mortality and Clinical Signs in Dams

One death at the HD on day 8 of gestation was attributed to treatment. Observations of treatment-related ataxia, decreased activity, pale extremities, red material around vagina, and unkempt appearance were made only in HD dams.

3. Dam Body Weight

Dose-related decreases in maternal BW (E16 means -4 and -6% at MD and HD, respectively, compared to C) and in BW gain (-30 and -40% at MD and HD, respectively, over days 5-15; **Table VI.4**) were seen in treatment groups compared to controls.

Table VI.4: Maternal Body Weight Gain (mean grams ± SE)

Day of Gestation	Dose (mg/kg)			
	0	20	100	500
6 to 15	53.4 ± 1.21	48.5 ± 1.71	36.7* ± 1.66	31.9* ± 2.10
0 to 20	145.9 ± 2.89	139.4 ± 3.25	128.0* ± 4.21	125.8* ± 3.43

* Significantly different from control, $p \leq 0.05$

4. Maternal Serum Drug Levels

In a pilot study, serum concentrations of topiramate were determined prior to dosing and 1-1.5 hr postdosing on days 12-15 of gestation in pregnant rats (N=4/group) dosed with 0, 200, 400, 600, or 800 mg/kg. Measurable concentrations of drug were observed in all predose samples; postdose levels increased approximately dose-proportionally(**Table VI.5**).

Table VI.5: Serum Topiramate Concentrations in Pregnant Rats

Dose (mg/kg)	Mean Concentration (ug/ml)	
	Predose	Postdose
200	7.0 ± 1.6	97.3 ± 17.7
400	6.0 ± 2.7	168.6 ± 30.1
600	12.4 ± 10.1	279.7 ± 129.2
800	35.1 ± 33.3	369.5 ± 70.6

5. Reproductive Data

a) Resorptions were increased (2X) at the HD compared to C, but the mean value (0.86/dam) was within the historical range. Implants also may have been somewhat increased at HD.

b) Fetal BWs were decreased at all doses compared to C (7, 15, 25% at LD, MD, & HD, respectively; **Table VI.6**).

Table VI.6: Fetal Body Weight (mean grams ± SE)

	Dose (mg/kg)			
	0	20	100	500
Male	3.32 ± 0.03	3.08* ± 0.04	2.80* ± 0.06	2.47* ± 0.04
Female	3.44 ± 0.04	3.17* ± 0.04	2.88* ± 0.05	2.56* ± 0.04
Combined	3.20 ± 0.04	2.98* ± 0.04	2.70* ± 0.06	2.37* ± 0.04

* Significantly different from control, $p \leq 0.05$

6. Fetal Evaluations

All fetuses examined for external defects; half each examined for skeletal and visceral (Wilson's) defects.

a) Incidences of specific limb malformations typically associated with CA inhibitors (ectrodactyly, micromelia, limb agenesis) were increased at the HD (**Table VI.7**). The anomalies seen in topiramate-exposed fetuses were predominantly right-sided; however, it was not clear that the the postaxial forelimb was primarily involved (right-sided postaxial forelimb preferentially affected by other CA inhibitors). One case of polydactyly (not normally produced with CA inhibitors) was also found at the HD.

b) *In addition to the HD limb defects, single fetuses with micromelia and/or* ectrodactyly were observed in the LD and MD groups. Micromelia and ectrodactyly were seen in 1 LD fetus displaying the "short thick body syndrome," which also occurred in 1 C fetus (without limb defects). Micromelia in 1 MD fetus was atypical in that it involved all four limbs and was apparently also associated with this syndrome. The sponsor questioned whether these cases were treatment-related; however, short thick body syndrome does not normally include limb effects.

c) Skeletal variations, including reduced/unossified skull bones, sternebrae, vertebrae, fore-/hindpaws, pelvis, and ribs, were increased in MD and HD litters compared to C. Reduced ossification was particularly pronounced in the fore- and hindpaws, which were equally affected.

Table VI.7: Summary of Fetal Malformations in Rats (Study #1)

	Dose (mg/kg)			
	0	20	100	500
No. of litters examined	49	25	23	22
No. of fetuses examined externally	648	338	288	318
No. of fetuses examined viscerally	222	114	94	107
No. of fetuses examined skeletally	426	224	194	211
Malformations in fetuses (litters)				
Anophthalmia	0	2 (2)	0	0
Arthrogryposis	3 (2)	0	0	0
Brain ventricles malpositioned	0	1 (1)	0	0
Cleft palate	0	0	1 (1)	0
Ectrodactyly	0	1 (1)	0	18 (4)
Enceohalocele	0	1 (1)	0	0
Heart malformation	0	1 (1)	0	0
Limb agenesis	0	0	0	3 (1)
Microphthalmia	0	2 (2)	0	0
Micrognathia	0	0	1 (1)	0
Micromelia/malformed long bones	0	1 (1)	1 (1)	11 (3)
Polydactyly	0	0	0	1 (1)
Short thick body syndrome	1 (1)	1 (1)	1 (1)	0
Skull bone absent	0	0	1 (1)	0
Spina bifida	0	1 (1)	0	0
Tail Agenesis	0	1 (1)	0	0
Vertebral malformation	2 (2)	0	0	0
Total fetuses (litters) with malformations	5 (4)	4 (4)	1 (1)	21 (6)

E) TERATOLOGY STUDY IN RATS (Kyowa Hakko Study (243794:2; GLP; Vol. 35)

1. Treatment

35 pregnant females/group were dosed with 0, 0.2, 2.5, 30 or 400 mg/kg, by gavage, on days 6 through 15 of gestation. C-sections were performed on 23 dams/group at 20 days of gestation. Remaining dams (12/group) were allowed to deliver and raise offspring to weaning. Selected F1 were maintained for behavioral testing and mating.

Strain: Crj:CD(SD) from Charles River Japan
Drug lot #: 9006522

2. Mortality and Clinical Signs in Dams

Ataxia, hypoactivity, urogenital staining, depilation, and diarrhea were observed in HD dams. No treatment-related mortality was seen.

3. Dam Body Weight

Decreased BW (7% on E20), BW gain (20% over days 0-20, 40% over days 6-15), and food consumption were observed in HD dams.

4. C-Section Data

a) Reproductive and Fetal Parameters

Fetal BWs were decreased at 30 (10%) and 400 mg/kg (25%) compared to C.

b) Fetal Evaluations

All fetuses examined for external defects, 1/3 for visceral (Wilson's and Nishimura's methods) and 2/3 for skeletal defects.

i. Right forepaw ectrodactyly was seen at the HD only, in 6 fetuses from 4 litters (**Table VI.9**).

ii. Total visceral anomalies were increased in a dose-related manner in all treated groups (4, 5, 7, 9, and 15 litters affected at 0, 0.2, 2.5, 30, and 400 mg/kg, respectively; statistically significant at HD). This was primarily due to a D-T increase in the incidence of thymic remnant in the neck in all treatment groups as well as to increased incidences of hydronephrosis and kinked ureter at 30 and 400 mg/kg. Although classified as malformations in this study, all 3 of these visceral anomalies are usually considered variations, and their increased incidence probably reflects treatment-related developmental delay.

iii. Total numbers of litters and fetuses with skeletal variations were increased at doses of 2.5 mg/kg and greater [16 (46), 14 (44), 19 (53), 21 (84), and 22 (146) litters (fetuses) affected at 0, 0.2, 2.5, 30, and 400 mg/kg, respectively; statistically significant at HD]. Specifically, skull ossification was delayed at 30 and 400 mg/kg, and vertebral anomalies (absence, deformity, splitting, supernumerary, abnormal alignment) and rudimentary lumbar ribs were increased at doses of 2.5 mg/kg and greater. Rib shortening and full lumbar ribs were observed only at the HD.

Table VI.8: Summary of Fetal Malformations in Rats (Study #2)

	Dose (mg/kg)				
	0	0.2	2.5	30	400
No. of litters examined	22	22	23	22	23
No. of fetuses examined externally	317	328	323	328	327
No. of fetuses examined viscerally	106	109	105	109	110
No. of fetuses examined skeletally	211	219	218	219	217
Malformations in fetuses (litters)					
Abnormal origin of rt subclav artery	0	0	0	0	1 (1)
Anal atresia	0	0	1 (1)	0	0
Anencephaly	0	0	1 (1)	0	0
Ectrodactyly	0	0	0	0	6 (4)
Edema	0	0	1 (1)	0	0
Encephalocele	0	0	1 (1)	0	0
Hydronephrosis	0	1 (1)	0	2 (2)	4 (4)
Hypoplasia of uterus	1 (1)	0	0	0	0
Interruption of aortic arch	0	0	1 (1)	0	0
Kinked ureter	4 (2)	1 (1)	2 (2)	4 (4)	5 (4)
Left umbilical artery	0	2 (2)	1 (1)	1 (1)	0
Omphalocele	0	0	1 (1)	0	0
Renal hypoplasia	0	0	1 (1)	0	0
Sternebral fusion	0	0	1 (1)	0	0
Thymic remnant in the neck	1 (1)	4 (4)	8 (5)	9 (7)	22 (14)
Transposition of great vessels	0	0	1 (1)	0	0
Vertebral anomaly	0	3 (3)	0	0	1 (1)
Vestigial tail	1 (1)	0	0	0	0
Total fetuses (litters) with malformations	7 (5)	10 (8)	12 (7)	13 (9)	33 (16)
No. fetuses (tters) w/					
External malformations	1 (1)	0 (0)	1 (1)	0 (0)	6 (4)
Visceral malformations	6 (4)	7 (5)	11 (7)	13 (9)	27 (15)
Skeletal malformation	0 (0)	3 (3)	1 (1)	0 (0)	1 (1)

5. Delivery Data

a) Reproductive and Offspring Parameters

A very slight increase in dead pups was seen on day 0 in MD and HD litters (5 in each versus 1 in C).

b) Pup Observations

i. Four cases of ectrodactyly of the right forepaw were observed at the HD (3 litters).

ii. BW gain and food consumption were decreased after weaning through week 11 at 30 and 400 mg/kg (final wts 5-10% below C in HD).

iii. Incisor eruption was delayed in HD offspring. No effects on tests of vision, hearing, and equilibrium during postnatal development.

iv. A tendency toward dose-dependent increases in ambulations was seen in males, but no other changes were observed in open field tests. There were no treatment-related effects on water maze learning.

v. No effects on reproductive capacity.

vi. Decreased kidney weights were seen in HD offspring compared to C at weaning necropsy, but no differences in degree of ossification were

observed among litters. In pathology examinations of HD males used in the reproductive capacity study (attempt to explain persistent growth deficit), slightly decreased WBCs and serum bilirubin and unilaterally decreased kidney weights were observed in the treated animals compared to controls.

F) TERATOLOGY STUDY IN RABBITS (McNeil Study (238725:2; GLP; Vol. 37)

1. Treatment

24 pregnant females were dosed with vehicle and 16 females/group dosed with 20, 60, or 180 mg/kg, by gavage, on days 6 through 18 of gestation. C-sections were performed on day 29 of gestation.

Strain: New Zealand White
Drug lot #: 8420100

2. Mortality and Clinical Signs in Dams

a) A T-R increase in mortality was observed, with 8, 0, 25, and 50% mortality in the C, LD, MD, and HD groups, respectively.
b) There was a dose-related increase in clinical signs, including hypoactivity, dyspnea, bloody discharge, and decreased defecation.
c) Abortion was increased at the MD and HD; 1, 1, 3, and 2 does aborted in the C, LD, MD, and HD groups, respectively.
d) Postmortem findings in HD animals included single incidences of dehydration, pyelonephritis, renal calculi and depressed kidney foci.

3. Dam Body Weight

Dose-related maternal BW loss was observed across treatment groups during the treatment period (-17% in HD versus +2% in C over days 6-18). During the post-treatment interval (days 19-29) weight gain in treatment groups was increased compared to C (HD BW 95% of C on day 29).

4. Litter Data

a) Reproductive and Litter Parameters (**Table VI.9**)

i. There was a dose-related reduction in the number of does with viable fetuses at C-section, due to death and abortion (19, 12, 10, and 6 viable litters in C, LD, MD, and HD, respectively).
ii. Resorptions and postimplantation loss were increased in MD and HD litters, and viable fetuses were decreased in HD litters (-50% compared to C, see below).
iii. Although treatment should have started around the time of implantation, corpora lutea were decreased and preimplantation loss was increased at the HD.
iv. Female fetal BWs were dose-dependently decreased (-10% at HD).

Table VI.9: Reproductive and Litter Parameters in Rabbits (Study #1)

	Dose (mg/kg)			
	0	20	60	180
Maternal parameters				
No. pregnant	21	13	16	16
No. dead	2	0	4	8
No. delivered early	1	1	3	3
No. w/ viable litters	19	12	10	6
Corpora lutea/doe (mean ± SE)	10.0 ± 0.4	10.8 ± 0.4	10.0 ± 1.0	7.7 ± 1.5
Implant sites/doe	9.4 ± 0.4	9.5 ± 0.8	9.5 ± 0.8	6.7 ± 1.3
Live fetuses/doe	8.4 ± 0.4	8.8 ± 0.9	7.9 ± 0.6	4.3 ± 1.0
Resorptions/doe	1.0 ± 0.3	0.7 ± 0.4	1.6 ± 0.5	2.3 ± 1.1
% Preimplantation loss	6.2 ± 1.5	12.6 ± 6.2	4.8 ± 2.6	16.1 ± 7.9
% Postimplantation loss	9.5 ± 3.2	7.6 ± 5.3	14.9 ± 3.9	28.1 ± 11.5
Fetal Parameters				
Body weight (gm, mean ± SE)				
Combined	40.3 ± 1.4	38.6 ± 3.2	38.3 ± 2.4	38.8 ± 3.7
Male	41.0 ± 1.3	37.1 ± 2.5	39.1 ± 2.6	40.6 ± 2.7
Female	40.2 ± 1.5	38.6 ± 3.4	37.5 ± 2.3	35.8 ± 4.9

b) Fetal Evaluations (**Table VI.10**)

All fetuses were examined for external, skeletal and visceral (Staple's method) defects.

i. There appeared to be small non-D-R increases in total malformations and incidences of skeletal (rib and vertebral) defects at the LD and HD compared to C, but too few litters and fetuses were examined at the HD to make a valid assessment of teratogenicity.

ii. No obvious treatment effect on variations, but above problem also applies here.

Table VI.10: Summary of Fetal Malformations in Rabbits (Study #1)

	Dose (mg/kg)			
	0	20	60	180
No. of litters examined	19	12	10	6
No. of fetuses examined externally	160	106	79	26
No. of fetuses examined viscerally	160	106	79	26
No. of fetuses examined skeletally	160	106	79	26
Malformations in fetuses (litters)				
Fused ribs	0	2 (2)	0	2 (2)
Hydranencephaly	1 (1)	0	0	0
Hydrocephalus	0	0	0	1 (1)
Major vessel anomaly	0	1 (1)	0	0
Rib anomaly	0	1 (1)	0	0
Tail agenesis	0	0	0	1 (1)
Umbilical hernia	0	0	0	1 (1)
Vertebral anomaly	0	2 (2)	0	2 (2)
Total fetuses (litters) with malformations	1 (1)	4 (3)	0	3 (3)

G) TERATOLOGY STUDY IN RABBITS (Kyowa Hakko study, 243908:1; GLP; Vol. 37)

1. Treatment

15 presumed pregnant females/group were dosed with 0, 10, or 35 mg/kg and 20 females were dosed with 120 mg/kg, by gavage, on days 6 through 18 of gestation. C-sections were performed on day 28 of gestation.

Strain: Japanese white rabbits, Kbl:JW,SPF
Drug lot #: 9006522

2. Fo Mortality, Clinical Signs, and Necropsy

a) Mortality was increased by treatment, with 2/13 and 7/16 dead in MD and HD groups, respectively. Abortion was seen in 1 MD and 3 HD does.
b) Signs increased at the HD included ataxia, hypoactivity, bloody discharge, and diarrhea.
c) Postmortem findings in HD animals included hemorrhage, ulcers, and gi blood clots in 3 cases, hyperemia of the lungs in 1 case, discoloration of the liver in 2 cases, and discoloration of the kidneys in 1 case.

3. Fo Body Weight

Maternal BW loss was seen in MD and HD groups during the treatment period (-2 and -[illegible]%, respectively, versus +4% in C); however, there were no differences in day 28 BWs.

4. Reproductive and Fetal Parameters (**Table VI.11**)

a) Resorptions/implantations (early and late) were increased and live fetus/implants decreased at the MD (slight) and HD (significant).
b) Fetal BWs were decreased at the MD and HD (no D-R).

Table VI.11: Reproductive and Litter Parameters in Rabbits (Study #2)

	Dose (mg/kg)			
	0	10	35	120
Maternal parameters				
No. pregnant	14	13	11	11
Corpora lutea/doe (mean ± SD)	11.8 ± 1.8	10.8 ± 2.7	12.8 ± 1.7	13.4 ± 1.4
Implant sites/doe	8.3 ± 2.5	7.5 ± 3.0	11.3 ± 1.7	10.1 ± 2.9
% to corpora lutea	71.2 ± 21.7	69.3 ± 21.7	88.9 ± 14.3	75.7 ± 19.8
Live fetuses/doe	7.8 ± 2.5	7.2 ± 2.8	10.0 ± 2.3	6.7 ± 3.5
% to implants	94.1 ± 8.8	96.5 ± 5.7	88.5 ± 13.7	59.8 ± 29.7
Percent early death/implants	4.2 ± 8.5	3.5 ± 5.7	5.9 ± 7.1	16.6 ± 29.1
no. affected litters	3	4	5	7
Percent late death/implants	1.7 ± 4.4	0.0 ± 0.0	5.6 ± 10.3	13.6 ± 18.2
no. affected litters	2	0	3	6
Fetal Parameters				
Body weight (gm, mean ± SD)	36.1 ± 6.9	37.0 ± 5.4	32.5 ± 4.1	33.0 ± 5.0

5. Fetal Evaluations (Table VI.12)

All fetuses were examined for external, skeletal and visceral defects.

a) Total malformations and incidences of visceral (abnormal vertebral and thoracic arterial origin) and skeletal defects (thoracic and lumbar vertebral malformations, absence of ribs, branched ribs) were increased at the HD (small increase at MD). Cleft palate was found in 4 fetuses from 1 HD litter.

b) No treatment-related effects on skeletal variations were seen.

Table VI.12: Summary of Fetal Malformations in Rabbits (Study #2)

	Dose (mg/kg)			
	0	10	35	120
No. of litters examined	14	13	11	10
No. of fetuses examined externally	109	94	110	74
No. of fetuses examined viscerally	109	94	110	74
No. of fetuses examined skeletally	109	94	110	74
Malformations in fetuses (litters)				
Blood vessel anomaly	5 (4)	1 (1)	6 (5)	10 (8)
Cleft palate	0	0	0	4 (1)
Dextroposition of esophagus	0	0	1 (1)	0
Folded retina	0	0	0	1 (1)
Rib anomaly	1 (1)	1 (1)	2 (2)	8 (6)
Splenic hypoplasia	0	1 (1)	0	0
Sternebra anomaly	2 (2)	0	2 (2)	5 (2)
Thymic remnant	1 (1)	1 (1)	4 (2)	1 (1)
Umbilical hernia	0	0	0	1 (1)
Vertebral anomaly	1 (1)	1 (1)	3 (3)	13 (7)
Total fetuses (litters) with malformations	7 (5)	4 (4)	16 (9)	27 (10)
No. fetuses (litters) w/				
External malformations	0	0	0	5 (2)
Visceral malformations	5 (4)	3 (3)	11 (7)	11 (8)
Skeletal malformation	2 (2)	1 (1)	5 (5)	19 (9)

H) SEGMENT III STUDY IN RATS (A500807; GLP; Vol. 2.37)

1. Treatment

30 pregnant females/group were dosed with 0, 0.2, 4, 20, or 100 mg/kg, by gavage, from day 15 of gestation through day 20 of lactation.

Strain: Sprague-Dawley [Crl:CD BR VAF/Plus]

Drug lot #: 8806672

2. F0 Mortality

No deaths due to drug-treatment.

3. F0 Observed Signs

Ataxia and hypoactivity were noted at the HD during gestation, but there were no differences in drug-related signs during parturition and lactation.

3. F0 Body Weight

Decreased BW gain (-18%), and food consumption (-16%) were observed in HD group during the gestational treatment period, but there were no notable differences in BWs on day 21. Lactational BWs were comparable.

5. Delivery Data

There were no effects on gestation length or ratio of live pups on day 1/implant sites on day 21 of lactation.

6. F1 Mortality

Pup survival was not affected by treatment.

7. F1 Signs and Necropsy Observations

No treatment effects.

8. F1 Body Weight

Pup weights were decreased in 20 (slight) and 100 mg/kg (statistically significant) litters at birth; these deficits persisted throughout lactation (mean BW 6% & 14% below C, respectively, on P21).

VII. SUMMARY

PHARMACODYNAMICS

In initial screening (**Tables I.1-4**), topiramate was active against MES-induced seizures in mice (po MES ED50 = 43.8) and rats (po MES ED50 = 17.3) and was inactive or very weakly active against chemoconvulsant-induced seizures. Peak anticonvulsant activity was reached within 1 hr in the mouse and between 1 and 4 hr in rats. Duration of activity was >4 hr and >8 hr in mice and rats, respectively. The separation between anticonvulsant activity and neurotoxicity for topiramate compared favorably with prototype agents. No tolerance to the anti-MES effect was apparent after oral dosing for 14 days with twice the ED50, but tolerance development was seen after 5 days with four times the ED50. SKF-525A increased anticonvulsant activity in the mouse MES threshold test, suggesting that the parent compound is the active species. The anticonvulsant activity of topiramate was abolished by reserpine. This effect of reserpine has previously been reported with acetazolamide and other CA inhibitors. Topiramate (20 and 40 mg/kg ip) was also active against both tonic and absence-like seizures in spontaneously epileptic rats (SER) and potently inhibited sound-induced seizures in DBA/2 mice (ED50 = 8.6 mg/kg po). Activity against absence-like seizures in the SER was blocked by haloperidol pretreatment. Topiramate and acetazolamide were both less potent against MES in seizure-prone DBA mice with abnormally high brain carbonic anhydrase activity than in C57 mice with normal brain CA activity. In topiramate-treated mice (20 mg/kg ip), CA activity was inhibited by about 60% in whole blood, 90% in brain homogenates, and 80% in cytosol of whole brain tissue in both strains.

In mechanism-related studies, topiramate (10-200 uM) demonstrated inhibitory effects on spontaneous neuronal firing and on depolarization-induced sustained repetitive firing in cultured hippocampal neurons. The latter was indicative of state-dependent Na^+ channel blockade, an effect shared by phenytoin and carbamazepine. Ligand binding studies with topiramate revealed no interaction (at up to 10 uM) at various neurotransmitter and drug binding sites, including those associated with GABAA and NMDA receptors. However, patch-clamp studies showed that topiramate (1-200 uM) enhanced GABA-induced Cl^- currents and inhibited kainate-evoked inward currents in cultured neurons. It is thought that any or all of these properties (Na^+ channel blockade, potentiation of GABA receptor responses, inhibition of kainate/AMPA receptor activity) could account for or contribute to the anticonvulsant activity of topiramate.

A structural feature of topiramate, the side chain sulfamate moiety, suggested possible similarities to sulfonamide containing carbonic anhydrase (CA) inhibitors such as acetazolamide, which possess broad spectrum anticonvulsant activity. The anticonvulsant effect of acetazolamide is thought to be specifically related to noncompetitive inhibition of CA in the CNS. Topiramate was found to inhibit CA in all species tested, but was less potent than acetazolamide in the initial studies conducted with CA derived from erythrocytes and brain tissue. The relative potency of topiramate/acetazolamide ranged from 0.00004 (human erythrocyte) to 0.25 (mouse erythrocyte). There are known to be a number (at least 7) of isozymes of CA, however, and in subsequent studies topiramate appeared to have selective activity in inhibiting various CA isozymes from rats and humans. Topiramate was most potent in inhibiting a CA isozyme (CA II) which is thought to predominate in brain cytosol and myelin, as well as in the kidney cytosol. Topiramate had a Ki similar to that of acetazolamide for this particular isozyme in rats (0.05-0.08 vs 0.01 uM; **Table I.6**). There were marked species differences in topiramate's CA inhibitory activity, and based on Ki values for human and rat CA II, the drug was 100-times more potent in rats. Although these data suggest that the anticonvulsant action of topiramate could result from its inhibitory effect on one or more specific neural CAs, the relationship between these two pharmacologic properties of topiramate remains to be clarified. Another anticonvulsant drug under development in Japan, zonisamide, contains a sulfamoyl moiety and is also a weak CA inhibitor, but studies conducted by the developer led them to conclude that the anticonvulsant activity of the drug was probably not due to the CA inhibitory effects of the compound. The anticonvulsant activity of zonisamide was not abolished by reserpine pretreatment, in contrast to the results with topiramate and acetazolamide. The use of acetazolamide in human epilepsy is limited by

tolerance development, believed to be due to increased CA activity. Studies in rats showed that tolerance to the anti-MES effect of acetazolamide occurred after administration of a high dose (4X MES ED50) for 5 consecutive days (**Table I.5A**). No cross-tolerance to the anticonvulsant effect of topiramate was observed in acetazolamide-pretreated mice, however. When a similar dose of topiramate was administered for 5 days, tolerance to topiramate as well as cross-tolerance to acetazolamide was seen (**Table I.5B**). These results were interpreted as indicating that topiramate has multiple mechanisms of action, one of which is shared by acetazolamide; however, they could reflect the different isozyme inhibitory profiles of the two drugs.

Gastrointestinal and renal effects of potential toxicological importance were seen after administration of topiramate. All of these were thought to result from CA inhibition. Gastric acid and gastric juice secretion were inhibited in rats and dogs and mucosal irritation was produced in rats by both topiramate and acetazolamide. Compared to acetazolamide, topiramate was more potent in rats and less potent in dogs in producing these effects. Diuresis, urinary alkalinization, increased electrolyte (Na^+, K^+, $HCO3^-$) excretion, and metabolic acidosis were seen with iv administration of topiramate to rats. These effects were qualitatively similar to, but less pronounced than, those produced by an equimolar dose of acetazolamide (**Table I.7**). Topiramate, at doses up to 10 mg/kg iv, had a small cardiovascular pressor effect in dogs, but did not alter the ECG. MAP and renal blood flow were decreased by topiramate in rats.

ADME

Absorption and Pharmacokinetics

Absorption was rapid after oral administration of topiramate, with maximum serum or plasma concentrations in rats, rabbits, and dogs measured at 0.5-2 hr, 1.3-1.7 hr, and 0.6-4 hr, respectively. The absolute bioavailability of a single oral gavage dose of 30 mg/kg was close to 100% in rats, based on AUCs. After a single oral dose (30 mg/kg), topiramate plasma Cmax values for male (27.6 ug/ml) and female (33.8 ug/ml) rats were similar (**Table II.1**); but the half life was shorter in males (2.6 vs 4.7 hr), and the clearance was greater in males (3.6 vs 1.6 ml/min/kg). Dose-normalized AUCs were similar within sex after administration of single oral doses of 10 and 300 mg/kg to rats, indicating dose proportionality; but AUCs were 2.5-3-fold higher in females than in males (**Table II.2**). After administration of a single oral dose (60 mg/kg) to female rabbits, topiramate plasma Cmax, AUC, CL/F, and t1/2 values of 39.5 ug/ml, 212 ug•hr/kg, 4.8 ml/min•kg, and 2.9 hr, respectively, were determined (**Table II.4**). After administration of a single oral dose (40 mg/kg) to 2 male and 2 female dogs, combined sex topiramate plasma Cmax, AUC, CL/F, and t1/2 values of 45.4 ug/ml, 190 ug•hr/kg, 3.6 ml/min/kg, and 2.6 hr, respectively, were measured (**Table II.5**). Pharmacokinetic parameters after multiple dosing (8 or 14 daily doses) were not significantly different from those observed after a single oral dose in rats, rabbits and dogs, indicating that there was no appreciable accumulation or autoinduction at the doses tested.

Topiramate was rapidly absorbed after administration of single oral doses of 100-1200 mg to human volunteers, with mean peak plasma concentrations occurring between 1.4 and 4.3 hr. The increase in peak plasma concentrations was linear but not proportional with respect to dose. Mean t1/2 ranged from 18.7 to 23 hr and was not dose-dependent. Mean oral plasma clearance was inversely related to dose, ranging from 36.1 ml/min at the LD to 22.5 at the HD. The mean Vd was also reduced at higher doses, ranging from 58 to 38.5 L over the doses used. This effect is probably due to saturable binding to a low capacity binding site on erythrocytes. A linear and proportional increase in Cmax and AUC values was observed with once-daily dosing of 50 to 200 mg or twice-daily dosing of 50-100 mg for 14 days. No changes in plasma or renal clearance or t1/2 were evident after multiple dosing. Studies done with 14C-labeled drug indicated that topiramate was well absorbed, not significantly metabolized, and excreted mainly in the urine. Intersubject variability in plasma concentration was low, and unchanged topiramate represented 85% of total plasma radioactivity at 24 hr after dosing.

Tissue Distribution

Following oral administration of 14C-topiramate (10 mg/kg) to male rats, the highest levels of radioactivity were found in the gi tract, blood, liver, and kidneys. Brain concentrations were less than or equivalent to those in the plasma. Radioactivity was rapidly eliminated from all tissues so that amount were negligible by 48 hr. Tissue levels of radioactivity were similar in pregnant and nonpregnant female rats after an oral dose of 20 mg/kg of 14C-topiramate. Concentrations in the fetus were similar to, and declined in parallel with, maternal plasma levels (12-13 ug-equiv/g). Tissue levels were initially similar in male and female rats, but protracted elimination resulted in higher levels in females at 24 and 48 hr.

Metabolism

A total of 8 metabolites have been identified in various species (**Figure II.1**, page 17), and the metabolic pathways appear qualitatively similar in the mouse, rat, rabbit, dog, and human. In the mouse and rabbit, the only major pathway was hydrolysis of the 2,3-O-isopropylidene group (path C) to produce a diol metabolite (**4**). Topiramate is substantially metabolized by male rats and by male and female dogs. In both, the major pathways appear to be hydroxylations at the isopropylidene groups (A and B) to form two hydroxylated metabolites (1 & 2). Pathway C was also important, as significant amounts of metabolite 4 were formed in both species. While the same metabolites were found in female rats and in humans, they were quantitatively minor (<3% of administered radioactivity). Metabolite 2 was shown to have anticonvulsant activity, but was about 1/3 as potent as topiramate in mice. SKF-525A increased the anticonvulsant activity of topiramate in mice, indicating that topiramate is a substrate for cytochrome p450 in this species. However, when topiramate was incubated with human liver microsomes, no metabolites were detected.

Elimination

After oral administration of 14C-topiramate, the major route of elimination for total radioactivity and unchanged topiramate was via the kidney in mice, rats, rabbits, dogs, and humans. After administration of 14C-topiramate to various species, 69-99% of the dose was recovered in urine. Elimination was essentially complete within 96 hr in all but humans, who excreted 14% of the dose in the urine from 96 to 240 hr. Probenecid increased renal clearance of topiramate in female rats, indicating that the renal reabsorption is important. Male and female rats excreted 38% and 5%, respectively, of total radioactivity in the bile over 24 hr. In lactating rats, about 1.5% of administered radioactivity was excreted in the milk over 24 hr.

Protein Binding

Plasma protein binding (in vitro) was low in all species tested; 6-17% was bound in mouse, rat, rabbit, dog, monkey, and human plasma at concentrations ranging from 1-250 ug/ml (clinical range extends to about 30 ug/ml). Evidence of saturable binding was seen in mouse, monkey, and human plasma. A high affinity, low capacity binding site was identified in erythrocytes from all species studied. The proportion bound to erythrocytes was higher at low plasma concentration (< 4 ug/ml), resulting in an increase in the blood to plasma concentration. Thus, clearance may be reduced at low plasma levels.

Toxicokinetics

A separate toxicokinetics study was conducted to support the mouse carcinogenicity study. After administration of topiramate (20, 75, and 300 mg/kg) as a dietary admixture to CD-1 mice (3/sex/group) for 1 month, 24-hr plasma AUCs were 11.2, 55.1, and 225.3 ug•h/ml in LD, MD, and HD males, respectively, and 2.5, 12.6, and 133.2 ug•h/ml in LD, MD, and HD females, respectively. A toxicokinetic study was also conducted to support the 12-month rat toxicology study. After administration of topiramate

(10, 55, and 300 mg/kg) as a dietary admixture to Wistar rats (5/sex/group) for 3 months, 24-hr serum AUCs were 27.8, 144.9, and 356.4 ug•h/ml in LD, MD, and HD males, while corresponding female values were 79.3, 361.4, and 1083 ug•h/ml. An approximate serum Cmax of 168 ug/ml was measured in pregnant rats dosed with 400 mg/kg in a separate toxicokinetic study. In clinical trials, a steady-state 24-hr AUC of 475 ug•hr/ml was obtained with a topiramate dose of 400 mg bid in patients also receiving VPA, and Cmax values up to about 30 ug/ml have been obtained. These are said to be the highest values likely to be seen in epilepsy patients.

Enzyme Induction

After oral administration of topiramate at 750 mg/kg (gavage) for two weeks or at 10, 55, or 300 mg/kg (diet) for nine months, significant increases in cytochrome P450 content, 7-ethoxycoumarin O-deethylase (ECOD) activity, and morphine glucuronyltransferase activity were measured in microsomes from male and female rats. Benzo[a]pyrene hydroxylase activity, ethoxyresorufin O-deethylase (EROD) activity, and acetaminophen glucuronyltransferase activity were increased in females only. EROD activity specifically reflects induction of isozymes from the cytochrome P450IA subfamily. ECOD reflects induction of a broader spectrum of isozymes, including P450IA and IIB (phenobarbital-inducible) subfamilies. The observed induction was considered moderate or weak compared to known inducers, such as phenobarbital and β-Naphthaflavone, although no direct comparisons were made (**Table II.11**).

Drug Interactions

Pretreatment of female rats for 7 days with phenytoin (100 mg/kg/day) increased the metabolism of a single oral dose of 14C-topiramate (60 mg/kg), raising the percentage of urine radioactivity accounted for by metabolites from 18 to 53%. Pretreatment of male rats for 7 days with phenytoin increased the clearance of an oral dose of topiramate (30 mg/kg) from 5.4 to 15.9 ml/min/kg. Both results indicate induction of topiramate metabolism by phenytoin. In human liver microsomal preparations, topiramate inhibited the 4'-hydroxylation of S-mephenytoin and the 1"-hydroxylation of 1'R-bufuralol in CYP2D6 deficient microsomes, ie CYP2Cmeph inhibition, indicating a potential drug interaction between topiramate and phenytoin. In a clinical interaction study, phenytoin clearance decreased in some subjects when topiramate was introduced.

TOXICOLOGY

The following acute, subchronic and chronic studies were performed (dose in mg/kg):

A,B,C) Acute toxicity in mice, rats, and dogs
D) 3-month oral toxicity in rats (10, 90, 750)
E) 3-month oral toxicity with recovery in rats (10, 90, 750)
F) 12-month oral toxicity in rats (10, 55, 300)
G) 11-month oral toxicity with recovery in rats (>300 M,>450 F)
H) 3-month oral toxicity in dogs (10, 40, 150)
I) 12-month oral toxicity in dog (10. 30, 100)

Acute Toxicity

Acute toxicity was evaluated in mice, rats, and dogs. Oral LD50's ranged from approximately 2000 to 4000 mg/kg in rodents. Male mice were more sensitive than females, while the reverse was true in rats. Dogs appeared to be considerably more sensitive than rodents, and male dogs were more sensitive than females. Clinical signs in all species tested were primarily related to CNS effects, and included ataxia, decreased activity, tremors, and clonic convulsions.

Multiple-Dose Toxicity (**Table VII.1**)

Rat

Two 3-month studies were conducted with oral (gavage) doses of 10, 90, and 750 mg/kg. One HD animal died in each of the 3-month studies. CNS signs, such as ataxia, amyotonia, and decreased activity, were observed with a dose-related incidence in both studies, with loss of righting at the HD. BW gain was decreased at the MD and HD in both studies, and BWs were about 5-10% below controls in these dose groups at the end of treatment. Water consumption was increased in MD and HD animals in one study, and the effect persisted after the recovery period. Increased RBCs and decreased MCHC noted at the HD in one study were attributed to hemoconcentration. Decreased prothrombin time, primarily in HD females, was the only hematology finding in the second study. Clinical chemistry findings were similar in the two studies, consisting of decreased serum potassium and increased serum protein, BUN, bilirubin, cholesterol, and phospholipids in drug-treated groups, primarily at the HD. Increased urine pH and volume and decreased urine specific gravity were seen at the MD and HD in both studies, while urine electrolyte effects were variable in these dose groups. Clinical chemistry and urinalysis changes were shown to be reversible in the recovery study. Liver and kidney/urinary tract findings observed in both 3-month studies, primarily in MD and HD animals, included increased liver and kidney weights, centrilobular hepatocellular hypertrophy, and increased incidences of pyelonephritis and hyperplasia of the kidney and urinary bladder transitional epithelial. Kidney and urinary tract effects were more pronounced in females, and recovery was demonstrated for all but the bladder urothelial hyperplasia in the second study. The LD (10 mg/kg) in these two studies was a NOAEL.

A 12-month study was conducted with oral (diet) doses of 10, 55, and 300 mg/kg. No treatment-related mortality and no clinical signs were observed. Decreased BW gain was seen in females from all dose groups and in MD and HD males, with terminal BWs decreased by about 10 and 20% in HD males and females, respectively. Erythrocyte parameters (RBCs, HGB, HCT) were decreased in a dose-related manner in treatment group animals, with values falling below the normal range at the HD. Serum chloride was increased at all doses, and cholesterol was increased and triglycerides decreased at the MD and HD. Urinalysis was not performed. Liver weights were increased in HD females, and kidney weights were increased in MD females and HD males and females. Treatment-related morphological changes (based on D-R incidence and severity) were observed in the fundic stomach, kidney, ureter, urinary bladder, and liver. Alterations in the gastric mucosa were found in all treatment groups (**Table III.1**). These were described as a diffuse hyperplasia of the generative cell zone of the neck of the gastric gland, with an associated reduction in parietal cells (frank degeneration at HD) and increase in the foveolar epithelium. Similar stomach changes were associated with increased gastrin levels in a separate 11-month study using high doses of topiramate (>300 mg/kg), which also demonstrated the reversibility of these effects. No increase in enterochromaffin-like cells was observed in either study (ECL cell hyperplasia has been shown to precede the development of carcinoid tumors in chronic studies with some antisecretory agents), and the proliferative changes were not considered preneoplastic by a consulting pathologist. The renal and urinary tract effects observed in topiramate-treated rats in the 12-month study, primarily in MD and HD groups, included urolithiasis, focal mineralization of the proximal convoluted tubules, urothelial hyperplasia, and papillary duct epithelial hypertrophy. Urothelial hyperplasia was thought to be secondary to mineralization and calculus formation (**Tables III.2-3**) and was not considered preneoplastic. Centrilobular hepatocellular hypertrophy was also seen in MD and HD rats. No NOAEL was established in the 12-month study because of the stomach effects seen at the LD.

A separate toxicokinetics study was conducted using the same doses as those in the 12-month study. After administration of topiramate in the diet to Wistar rats (5/sex/group) for 3 months, 24-hr serum AUCs of 27, 145, and 356 ug•hr/ml were obtained for LD, MD, and HD males, respectively. The corresponding values for females were 79, 361 and 1083 ug•hr/ml.

Dog

A 3-month study was performed with oral (capsule) doses of 10, 40, and 150 mg/kg. No deaths and no clinical signs were observed. BW gain, food consumption, and food efficiency were decreased in MD males and HD males and females. Erythrocyte parameters (RBCs, HGB, HCT) were slightly decreased in all treatment groups (10-15% at HD). Platelets and indices were increased slightly at the HD. There was no morphological evidence of extramedullary hematopoiesis, increased hemosiderin deposition, or bone marrow depletion. BUN and alkaline phosphatase values were increased and transaminases and total protein were decreased in treated animals, primarily at the HD. Serum potassium was decreased and chloride increased in treatment group dogs. Urine pH was increased and specific gravity decreased in all treatment groups. Liver weights were increased in MD females and in HD males and females, but no histopathology was observed. The NOAEL was 10 mg/kg. Serum concentrations measured at one hour postdose in the 14th week of the study were 9, 18, and 106 ug/ml in LD, MD, and HD males, respectively, and 8, 19, and 130 ug/ml in LD, MD, and HD females, respectively.

In a 12-month study with oral (capsule) doses of 10, 30, and 100 mg/kg, effects were similar to those observed in the 3-month study. There were no treatment-related deaths. The only treatment-related clinical observation was emesis, seen at all dose levels. BW gain was decreased 10-20% at the HD. Small D-R decreases in RBCs, HCT, and HGB were seen throughout treatment (10-20% at HD). One HD female (3234) had more severely depressed HCT, HGB, MCH, & MCV values with increased platelets and MCHC. Generally, RBC indices and platelets were somewhat increased compared to controls. Alkaline phosphatase, chloride, and cholesterol were increased and potassium, transaminases and total protein decreased in treatment group dogs; however, changes were not pronounced. Urine pH and volume were increased in all treatment groups, and urate and calcium crystal were increased at the MD and HD. Liver weights were increased and kidney and spleen weights slightly decreased at the HD. No histopathology was observed. The LD was considered a NOAEL.

CARCINOGENICITY **(Table X.2)**

The following studies were performed (doses in mg/kg):

A) 2-year carcinogenicity study in mice (20, 75, 300)
B) 2-year carcinogenicity study in rats (20, 45, 120)

Mouse

CD-1 mice were administered topiramate doses of 0 (C I), 0 (C II), 20, 75, or 300 mg/kg in the diet for 53 (10/sex/group) or 93-95 weeks (60/sex/group). Study duration was shortened due to high mortality (50-70%). There were no group differences in survival for males, but mortality was increased in HD females compared to one of the two control groups. BWs were slightly (<10%) decreased in HD groups compared to controls throughout the study. No treatment-related clinical signs were noted.

Incidences of the following morphologic changes were increased in treated mice at the 1-year interim sacrifice: hepatocellular hypertrophy in males, dilated gastric glands in males and females, lymphocytic infiltration and amyloidosis of the gastric mucosa in females, renal tubular dilatation and urinary bladder distension in females.

At study termination, gastrin levels were found to be slightly elevated in HD females, and treatment-related non-neoplastic morphologic changes were seen in the liver, stomach, kidneys, and urinary bladder, primarily of MD and HD group mice. These included (1) hepatocellular hypertrophy (MD, HD) and hyperplasia (HD males); (2) hyperplasia of the mucosal generative cell zone (MD, HD), lymphocytic infiltration (MD, HD), and focal hyperplastic diverticula (HD males) in the body of the stomach; (3) gross

kidney changes, renal pelvic dilatation, and chronic pyelonephritis or glomerulonephritis (all in HD females); and (4) urinary bladder calculi (HD females), chronic or hyperplastic cystitis (HD females), focal mucosal or stromal bladder hyperplasia (HD males; MD & HD females), and dilatation of the lumen of the urinary bladder (females at all doses).

The incidence of urinary bladder tumors was increased in HD males (4/60 vs 0/59 & 1/59 in controls) and in females from all treatment groups (3/60, 2/60, & 9/60 in LD, MD, HD, respectively, vs 1/60 & 1/60 in controls). Statistical significance was reached in HD males and females. There was no evidence of metastasis. The predominant tumor type, first diagnosed as a leiomyosarcoma, was later examined by consultant pathologists and determined to be histomorphologically unique to mice (see Evaluation). Single incidences of other types of bladder tumors (transitional cell carcinoma, papilloma, hemangiosarcoma) were found only in HD females (see **Table IV.1**). In addition, increased (but low) incidences of the following tumor types were observed in treated mice: squamous cell carcinoma of the (fore)stomach in males (0, 1, 1, & 2 in C II, LD, MD, & HD, respectively), uterine hemangioma (1, 0, 2, & 3 in C II, LD, MD, & HD) and leiomyosarcoma (2 HD), cervical leiomyoma (3 MD, 2 HD), lung bronchiolo-alveolar carcinoma in females (0, 2, 1, 2 in C II, LD, MD, & HD), ovarian hemangioma (1 HD), splenic hemangiosarcoma (1 LD male, 1 MD & 1 HD female), malignant meningioma (1 HD male), hepatocellular carcinoma in females (1 HD), and renal tubular carcinoma (1 HD male). None of these were significant in the FDA statistical analysis.

At the HD used in this study, 24-hr plasma AUCs measured in satellite groups were 225 and 133 ug•hr/ml in males and females, respectively. Plasma AUCs as high as 475 ug•hr/ml have been measured in clinical trials in patients receiving 400 mg/kg bid of topiramate.

Rat

Wistar rats (50/se/group) were administered topiramate doses of 0 (C I), 0 (C II), 20, 45, or 120 mg/kg in the diet for 104-105 weeks. Survival was not affected by drug treatment and was adequate for study validity (>50%). BWs were decreased in HD males (terminal mean 7% below C II) and in MD and HD females (final means 6 & 21% below C II). Serum gastrin levels were increased in HD males at 1 year and in MD and HD males and HD females at the end of treatment. No drug-related clinical signs were noted.

At study termination, treatment-related non-neoplastic morphologic changes were seen in the liver, stomach, kidneys, and urinary bladder. Findings included (1) hepatocellular hypertrophy (males at all doses, MD & HD females) and vacuolization (MD & HD females), and increased eosinophilic-cell foci (HD males, females at all doses); (2) hyperplasia of the generative cell zone of gastric glands in the body of the stomach, with an associated decrease in parietal cells and inflammatory cell infiltration, and hyperplasia of the limiting ridge between the forestomach and fundus (all at MD & HD); (3) renal calculi and hyperplasia of the pelvic and papillary urothelium (HD males; females at all doses); and (4) calculi (HD), mucosal hyperplasia (HD), and dilatation of the urinary bladder (MD & HD males).

There were no statistically significant increases in tumors in rats. Mucosal papillomas were found in the urinary bladders of 1 MD and 1 HD male and 1 HD female, possibly secondary to chronic irritation induced by calculi in the bladder lumen. In addition, a urinary bladder leiomyosarcoma was found in 1 MD female, a renal tubular carcinoma in 1 HD female, and a squamous cell carcinoma of the forestomach in 1 HD female.

Blood levels of topiramate were not determined for this study, but in satellite toxicokinetics groups for the 12-month rat toxicology study, administration (diet) of topiramate to Wistar rats at doses of 10, 55, and 300 mg/kg for 3 months produced 24-hr serum AUCs of 28, 145, and 356 ug•hr/ml, respectively, in males, with corresponding values of 79, 361, 1083 ug•hr/ml in females.

GENETIC TOXICITY

Topiramate was tested for mutagenicity in bacteria and in mouse lymphoma cells *in vitro*, for effects on unscheduled DNA synthesis in rat hepatocytes *in vitro*, and for clastogenicity in human lymphocytes *in vitro* and in rat bone marrow *in vivo*. Although there was a slight increase in chromosomal aberrations in the rat bone marrow test (**Table V.1**), topiramate was considered negative in all of these assays.

REPRODUCTIVE TOXICOLOGY

The following studies were performed (oral doses in mg/kg):

A) Segment I in male rat (0.2, 8, 25, 100)
B) Segment I in female rat (0.2, 8, 25, 100)
C) Segment II in mouse (20, 100, 500)
D) Segment II in rat (20, 100, 500)
E) Segment II in rat (0.2, 2.5, 30, 400)
F) Segment II in rabbit (20, 60, 180)
G) Segment II in rabbit (10, 35, 120)
H) Segment III in rat (0.2, 4, 20, 100)

Segment I Studies

Males were dosed with 0.2, 8, 25, or 100 mg/kg po for 70 days prior to mating and for 30 days after initiation of mating. BW gain was slightly reduced at 25 and 100 mg/kg, but no clinical signs were observed. There were no effects on fertility. Apparent effects on testes weight (increased), time to mating (increased), and postimplantation loss on day 13 of gestation (increased) were observed. Changes were small and not considered toxicologically relevant by the sponsor (no testes effects were noted in chronic studies). There were no treatment-related effects on reproductive parameters in pregnant females allowed to deliver naturally.

Females were dosed with 0.2, 8, 25, or 100 mg/kg po from 14 days prior to mating until termination on either day 13 of gestation or Day 21 of lactation. BW gain was reduced at 8 mg/kg or greater only during the first week of treatment, ie, prior to cohabitation. There were no treatment-related clinical signs. No effects on mating, pregnancy, or fecundity were observed. At 13 days of gestation, corpora lutea and implants were slightly decreased and postimplantation loss was slightly increased at 25 and 100 mg/kg (but <10% loss). No effects on gestation length, implants, litter size, or offspring viability were seen in dams allowed to deliver, but a persistent weight deficit was noted in HD pups.

No gross anomalies were observed in either study. Reproductive organs were not examined histologically in these studies.

Segment II Studies

Mice were dosed with 20, 100, or 500 mg/kg po on days 6 through 15 of gestation, and C-sections were performed on day 18. BW gain of pregnant females during gestation was decreased by about 20% in the HD group, but no other evidence of maternal toxicity was noted. Small increases in intrauterine death were seen in MD and HD litters, including 1 total litter loss at the HD. Fetal weights were decreased (4 & 10%, respectively) and numbers of small fetuses (<1 g) were increased at the MD (slight) and HD. Total malformations were increased at all doses. Malformed fetuses were found in 8.3, 25, 35, and 32% of C, LD, MD, and HD litters, respectively, and the number of malformed fetuses increased dose-dependently (**Table VI.3**). There was not a good dose-response of affected litters for any specific malformation (probably due to intrauterine death), but the litter incidence of craniofacial malformations was increased

in all treated groups. Cleft palate was observed in 0.37, 0.72, 1.40, and 2.54 percent of fetuses examined in the C, LD, MD, and HD groups, respectively. The MD and HD incidences exceeded the historical control range for this defect. Ablepharon was observed in 0, 0.72, 0.70, and 1.27 percent of C, LD, MD, and HD group fetuses, respectively. All treatment group incidences exceeded the historical control range for this defect. Single incidences of exencephaly were seen at the MD and HD only. Limb defects were found in single fetuses at the LD and MD (limb reductions at LD only, but CD-1 is considered a resistant strain for induction of these defects by CA inhibitors). Skeletal variations were increased in HD litters.

Rats were dosed with 20, 100, or 500 mg/kg po on days 6 through 15 of gestation, and C-sections were performed on day 20. Dose-related decreases in maternal BW gain were seen during the dosing period (-10, 30, & 40% in LD, MD, & HD, respectively). Clinical signs (ataxia and hypoactivity) were noted at the HD only, and one HD death was attributed to treatment. Fetal BW means were 7, 15, and 25% below controls at the LD, MD, and HD, respectively. Incidences of specific limb malformations typically associated with CA inhibitors (ectrodactyly, micromelia, limb agenesis) were increased in fetuses from the HD group (**Table VI.7**). The majority of topiramate-induced limb defects were right-sided, but it was not clear whether the postaxial forelimb was primarily involved (the postaxial right forelimb is preferentially affected by other CA inhibitors). One case of polydactyly (not normally produced by CA inhibitors) was also found at the HD. In addition to the HD limb defects, single fetuses with micromelia and/or ectrodactyly were observed in the LD and MD groups. Micromelia and ectrodactyly were seen in 1 LD fetus displaying the "short thick body syndrome," which also occurred in 1 C fetus (without limb defects). Micromelia in 1 MD fetus was atypical in that it involved all four limbs and was apparently also associated with this syndrome. Because of their association with what is thought to be a spontaneously occurring, genetically-determined syndrome, the sponsor questioned whether the limb malformations in the 2 lower dose group fetuses were treatment-related; however, this syndrome does not normally include limb effects. Skeletal variations (reduced/unossified skull bones, sternebrae, vertebrae, fore-/hindpaws, pelvis, and ribs) were increased in MD and HD litters. Reduced ossification was particularly pronounced in the fore- and hindpaws, which were equally affected.

In a second rat teratology study, rats were dosed with 0.2, 2.5, 30 or 400 mg/kg po on days 6 through 15 of gestation. Decreased BW (7% on E20), BW gain (20% over days 0-20, 40% over days 6-15), and food consumption were observed in HD dams. Clinical signs were increased in this group. An approximate Cmax of 168 ug/ml was measured in pregnant rats dosed with 400 mg/kg in a separate toxicokinetic study. In litters delivered by C-section on day 20, fetal BWs were decreased at 30 (10%) and 400 mg/kg (25%) compared to controls. Right forepaw ectrodactyly was seen at the HD only, in 6 fetuses from 4 litters (**Table VI.8**). Total visceral anomalies were increased in a dose-related manner in all treated groups. This was primarily due to a D-R increase in the incidence of thymic remnant in the neck in all treatment groups as well as to increased incidences of hydronephrosis and kinked ureter at 30 and 400 mg/kg. Although classified as malformations in this study, all 3 of these visceral anomalies are usually considered variations, and their increased incidence probably reflects treatment-related developmental delay. Skeletal variations (ossification delays, vertebral and rib variations) were dose-dependently increased at 30 mg/kg and greater, with full lumbar ribs seen only at the HD. In litters delivered naturally, four pups with right forepaw ectrodactyly (in 3 litters) were seen in the HD group. Incisor eruption was delayed in HD pups, and a post-weaning growth deficit was observed in pups from the 30 and 400 mg/kg litters. At necropsy, decreased kidney weights were found in HD pups, but no histopathology was reported.

Rabbits were dosed with 20, 60, or 180 mg/kg po on days 6 through 18 of gestation. Clinical signs (ataxia and hypoactivity), abortion and mortality were increased in MD and HD group does. Maternal BW loss during dosing occurred (D-R) in all treatment groups. At C-section on day 29, postimplantation loss was increased and viable fetuses decreased in MD and HD litters. There was a dose-related decrease in female fetal weights (HD 10% below C). Total malformations and skeletal defects appeared to be slightly increased at the LD and HD, but there were too few HD litters (6) and fetuses (26) to make a valid assessment of teratogenicity in this study. In a separate toxicokinetic study in non-pregnant rabbits, a

Cmax of 39 mg/kg and an AUC of about 200 ug·hr/ml were measured after administration of an oral dose of 60 mg/kg.

In rabbits dosed with 10, 35, or 120 mg/kg po on days 6 through 18 of gestation (second rabbit teratology study), decreased weight gain and increased mortality and abortion were seen in MD and HD does. Increases in treatment-related clinical signs were noted at the HD. There were 11 and 10 viable litters at C-section (day 28) in the MD and HD groups, respectively. Resorptions were increased and fetal BWs decreased in MD and HD litters (**Table VI.11**). Incidences of visceral (blood vessel anomalies) and axial skeletal defects were increased in HD fetuses (**Table VI.12**).

Segment III Studies

In rats dosed with 0.2, 4, 20, or 100 mg/kg po from day 15 of gestation through day 20 of lactation, clinical signs and decreased BW gain were seen in HD dams only during the gestational treatment period. There were no effects on gestation length, live pup/implants ratios, or pup survival; however, pup body weights were decreased at the two highest doses (mean BW 6 & 14% below C at 20 and 100 mg/kg, respectively, on P21; statistically significant at HD).

Table X.1: Summary of Subchronic and Chronic Toxicology Studies of Topiramate

Species/ Strain	Duration	No/ Sex/ Grp	Dose(mg/kg)/ Route/ Vehicle	Results
Rat/ Sprague-Dawley	3 months	15	10,90,750/ po gavage/ methylcellulose	10: no adverse effects 90: CNS signs; ↓ wt gain(♀); ↑ urine pH, volume; ↑ liver & kidney wts; hepatocellular hypertrophy; renal urothelial hyperplasia, microcalculi(♀) 750: CNS signs; ↓ wt gain (♀); ↓ serum K^+ (♀), ↑ protein, bilirubin, BUN, cholesterol; ↑ urine pH, volume; ↑ liver & kidney wts; hepatocellular hypertrophy; renal, bladder urothelial hyperplasia & microcalculi(♀) NOAEL = 10 mg/kg
Rat/ Sprague-Dawley	3 months + 4-week recovery	16	10,90,750/ Diet	10:↓ wt gain(♂) 90:↑ water consumption, urine pH, volume; hepatocellular hypertrophy; pyelonephritis; bladder urothelial hyperplasia (also seen in recovery grp) 750: CNS signs; ↓ wt gain; ↓ PT time (♀); ↓ serum K^+(♀), ↑ protein, bilirubin, BUN, cholesterol; ↑ water consumption; ↑ urine pH, volume; ↑ liver & kidney wts; hepatocellular hypertrophy; pyelonephritis, renal & bladder urothelial hyperplasia (bladder hyperplasia still seen after recovery) NOAEL = 10 mg/kg
Rat/ Wistar	12 months	25	10,55,300/ Diet	10: ↓ wt gain(♀); ↑ serum Cl^-; hyperplasia of gastric mucosa 55: ↓ wt gain; ↓ RBCs; ↑ serum Cl^-, cholesterol, ↓ triglycerides; ↑ kidney wts (♀); hyperplasia of gastric mucosa; hepatocellular hypertrophy; urothelial hyperplasia, urolithiasis, papillary duct epithelial hypertrophy 300: ↓ wt gain; ↓ RBCs; ↑ serum Cl^-, cholesterol, ↓ triglycerides; ↑ liver (♀) & kidney wts; gastric mucosal hyperplasia; hepatocellular hypertrophy; urothelial hyperplasia, urolithiasis, papillary duct epithelial hypertrophy, focal mineralization No NOAEL
Rat/ Wistar	11 months +recovery	21-26	M:>300 F:≥450/Diet	↓ wt gain; ↑ serum gastrin; gastric mucosal hyperplasia (reversal shown)
Dog/ Beagle	3 months	4	10,40,150/ po/gelatin capsule	10: ↑ urine pH 40: ↓ wt gain; ↓ RBCs; ↑ BUN, Cl^-, ↓ K^+, protein, SGPT, SGOT; ↑ urine pH; ↑ liver wts(♀) 150: ↓ wt gain; ↓ RBCs; ↑ BUN, Cl^-, ALP, ↓ K^+, protein, SGPT, SGOT; ↑ urine pH; ↑ liver wts NOAEL = 10 mg/kg
Dog/ Beagle	12 months	4	10,30,100/ po/gelcap	10: emesis; ↑ urine pH, vol 30: emesis; ↓ wt gain; ↓ RBCs; ↑ Cl^-, ALP, ↓ K^+, protein; ↑ urine pH, vol; ↑ liver wts 100: emesis; ↓ wt gain; ↓ RBCs; ↑ Cl^-, ALP, ↓ K^+, protein, SGPT, SGOT; ↑ urine pH, vol; ↑ liver wts, ↓ spleen wts No NOAEL

Table X.2: Summary of Carcinogenicity Studies of Topiramate

Species/ Strain	Duration	No/ Sex/ Grp	Dose(mg/kg)/ Route/ Vehicle	Results
Mouse/ CD-1	21 months	60	20,75,300/ po (diet)	Non-neoplastic 20: dilated urinary bladder lumen(♀) 75: gastric mucosal hyperplasia; hepatocellular hypertrophy; dilated urinary bladder lumen(♀), focal mucosal or stromal bladder hyperplasia(♀) 300: ↑ gastrin (♀); gastric mucosal hyperplasia, hyperplastic diverticula (♂); hepatocellular hypertrophy, hepatocellular hyperplasia (♂); dilated renal pelvis(♀), chronic pyelonephritis or glomerulonephritis(♀), renal calculi (♀); dilated urinary bladder lumen(♀), bladder calculi (♀), focal mucosal or stromal bladder hyperplasia(♀); Splenic hyperplasia (♂) Neoplastic Statistically significant increase in urinary bladder tumors in HD males and in females at all doses: C: bladder leiomyosarcoma (1♀, 1 ♂), leiomyoma (1♀) 20: urinary bladder leiomyosarcoma (3♀) 75: urinary bladder leiomyosarcoma (1♀), stromal polyp (1♀) 300: urinary bladder transitional cell carcinoma (1♀), hemangiosarcoma (1♀), papilloma (1♀), leiomyosarcoma ((6♀, 4♂)
Rat/ Wistar	24 months	50	20,45,120/ po (diet)	Non-neoplastic 20: hepatocellular hypertrophy (♂); renal calculi (♀), renal urothelial hyperplasia (♀) 45: hepatocellular hypertrophy, vacuolization (♀); gastric mucosal hyperplasia; renal urothelial hyperplasia (♀), renal calculi (♀); dilated urinary bladder lumen (♂) 120: hepatocellular hypertrophy, vacuolization (♀); gastric mucosal hyperplasia; renal, bladder calculi & urothelial hyperplasia; dilated urinary bladder lumen (♂) Neoplastic No statistically significant increases; following seen only in treated rats: 45: urinary bladder mucosal papilloma (1 ♂), bladder leiomyosarcoma (1♀) 120: urinary bladder mucosal papilloma (1 ♂, 1♀)

VIII. EVALUATION

The toxicity profile of topiramate has been adequately defined. Topiramate effects were generally consistent across studies and species, and carbonic anhydrase (CA) inhibition appears to be a common feature of many of these effects. Principle target organs were the stomach, kidney and urinary tract, and liver. There was also evidence of hematologic toxicity. Carcinogenicity and reproductive toxicity have also been adequately assessed. The major toxicological findings are discussed below.

Stomach

The gastric mucosal hyperplasia seen in topiramate-treated rodents (at doses as low as 10 mg/kg in the 1 year rat study) was attributed to hypergastrinemia or other changes in gastric physiology resulting from CA inhibition. Increased secretion of the hormone gastrin is known to occur in response to an elevated stomach pH, since gastric acidity is the negative feed back mechanism for inhibition of gastrin release from the G-cells. Both topiramate and acetazolamide were shown to decrease gastric acid secretion and raise stomach pH in rats, and high doses of topiramate increased serum gastrin in mice and rats. It is well established that hypergastrinemia has a generalized proliferative effect on the gastric mucosa. But, while gastrin is trophic to most gastric mucosa cells, topiramate administration reportedly produced specific changes in the cell populations of the glandular stomach. These were described as hyperplasia of the generative cell zone of the neck of the gastric gland with an associated increase in the foveolar epithelium and reduction in parietal cell mass. No increase in enterochromaffin-like (endocrine) cells was observed. Mucosal hyperplasia associated with hypergastrinemia has been reported after prolonged administration of various antisecretory agents (eg, H_2 blockers, proton pump inhibitors) to rodents, but in contrast to topiramate, these compounds induced the proliferation of ECL cells as well as mucous, parietal, and chief cells. ECL cell hyperplasia has been shown to precede the development of carcinoid tumors in lifetime rodent studies with these drugs. Thus, the hyperplastic condition observed in the stomachs of rodents exposed to topiramate does not appear typical of that associated with prolonged elevations in serum gastrin. Hyperplasia could also represent a regenerative response to the gastric mucosal ulceration produced in rats by topiramate, as well as acetazolamide. A similar proliferative condition of the gastric mucosa has been shown to develop following long-term treatment of rats with an ulcerogenic regimen of aspirin. The proliferative changes seen in the stomachs of topiramate-treated mice and rats were not considered dysplastic, and there was no evidence of progression to neoplasm in the 2-year studies. Although gastric acid secretion was decreased by topiramate in dogs, gastric hyperplasia was not observed in the dog toxicity studies, and neither increased serum gastrin nor gastric hyperplasia was seen in a small number of patients administered topiramate (200-500 mg) for 1-3 years.

Kidney and urinary tract

Several clinical pathology changes observed in the rat and dog studies were thought to reflect alterations in fluid and electrolyte levels resulting from diuresis, which was evident in the urinalysis results. The diuresis produced by topiramate (MD & HD in 3-month rat studies, all doses in dog studies) as well as the changes in urine and plasma composition (increased urine pH, Na^+, K^+, HCO_3^-; decreased plasma K^+, increased plasma Cl^-) were consistent with CA inhibition. CA inhibition was probably also involved in the pathogenesis of renal and urinary tract histological changes associated with topiramate. Calculus formation is a known side-effect of CA inhibitors (secondary to reduced urinary citrate and increased calcium), and the renal and bladder urothelial hyperplasia seen in topiramate-treated mice and rats was thought to be a response to urinary tract irritation produced by calculi or mineralization, as suggested by the association observed between these findings in rats (MD & HD in 3- and 12-month studies). Urinary alkalinization was considered a possible contributing factor. Both mechanical irritation and urine alkalinization have previously been shown to induce urothelial hyperplasia in rats, so this is a plausible explanation. In rat studies with acetazolamide and another (Merck) carbonic anhydrase inhibitor, early degenerative and inflammatory changes in the urinary bladder were followed by simple regenerative urothelial hyperplasia. These

changes, which were not observed in rabbits, dogs, or monkeys, were thought to be adaptive responses to the alterations in urine composition produced by CA inhibition. Similar effects have also been reported in mice. Although epithelial hyperplasia is a putative preneoplastic alteration, the proliferative changes seen in the urothelium of topiramate-treated mice and rats were not considered dysplastic. In a 3-month topiramate study in rats, where bladder urothelial hyperplasia was associated with doses as low 90 mg/kg, recovery had not occurred at 1 month after treatment withdrawal. Persistence of urothelial hyperplasia following withdrawal of treatment has been correlated with the development of bladder neoplasia in rats treated with cyclophosphamide; however the hyperplasia produced by this alkylating agent was characterized as atypical or dysplastic. Bladder urothelial hyperplasia in rats treated with acetazolamide (200 mg/kg for 4 weeks, Merck study) had only partially regressed after a 1-month recovery period, but the urothelium was normal 2 months after cessation of treatment. Other renal changes observed in topiramate-treated rats, such as papillary duct epithelial hypertrophy, focal mineralization of the proximal convoluted tubules, and pyelonephritis, have also been associated with administration of other carbonic anhydrase inhibitors. Urinary tract toxicity was seen at topiramate doses as low as 20 mg/kg in female rats in the 2-year study. Similar effects were not produced by the doses used in the dog studies, and the relevance of the rat findings to humans is uncertain. Urinary tract effects related to CA inhibition could result in significant adverse reactions in patients, however. A relatively high incidence of renal calculi terminated US clinical trials with zonisamide, a potential AED which is also a weak CA inhibitor.

Liver

Liver morphological findings and related clinical chemistry changes in rats and dogs were typical of those produced by hepatic enzyme inducers, and moderate induction of hepatic enzymes was demonstrated by topiramate in rat ADME studies. Increased liver weights and hepatocellular hypertrophy were consistently observed in MD and HD rats in the 3- and 12-month studies. Liver weights were increased at the MD and HD in both dog studies, with no apparent histological changes. These effects would not be expected to be clinically important.

Hematology

Small but significant reductions in RBC, HGB, and HCT values in topiramate-treated rats (MD & HD in 12-month study) and dogs (MD & HD in 3 and 12-month studies) were ascribed to hemodilution by the sponsor, but the mechanism was not elaborated on. The mild changes in erythrocyte parameters were not associated with morphological evidence of extramedullary hematopoiesis, increased hemosiderin deposition, or bone marrow depletion (increased extramedullary hematopoiesis was observed in the 2-year mouse study, but no other hematologic data is available for mice), and erythrocyte indices and platelet counts were usually increased or unchanged. Small treatment-related variations in hematologic values are often observed in preclinical toxicity studies and may result from a variety of physiological changes associated with prolonged exposure to high doses of drugs. Alterations in fluid and electrolyte balance or in food consumption and utilization have been shown to influence RBC values, so it is possible that the hematologic changes produced by topiramate were related to the renal or gi effects discussed above. Decreases in erythrocyte parameters have also been reported in preclinical studies with the antisecretory agents cimetidine and omeprazole in rats, which could indicate a common mechanism involving changes in gastric physiology. Chronic renal toxicity can decrease erythropoiesis, secondary to decreased erythropoietin; and a loss of gastric parietal cells, which are involved in the absorption of B12, could also affect erythrocyte production. Both factors may have played a role in the hematologic effects of topiramate in rats, although decreased RBC parameters were seen in dogs without obvious renal or stomach pathology. Concentrations up to 100 uM were not hemolytic in rat blood *in vitro*, but increased MCHC values in the dog studies suggest the possibility of hemolysis. Another hematologic finding of possible significance, in light of several thrombotic events reported in clinical trials, was a decrease in prothrombin times in one of the 3-month rat studies, primarily at the HD. *In vitro* testing with topiramate indicated no effects on blood coagulation or platelet aggregation, however, and similar effects were not seen in dogs.

It is unlikely that these topiramate findings have any clinical importance. Serious hematologic toxicity has been reported with therapeutic use of acetazolamide and other CA inhibitors, as with sulfonamide use in general; however, the dyscrasias associated with these drugs are thought to usually represent idiosyncratic reactions for which preclinical studies have poor predictive value.

Carcinogenicity

The incidence of urinary bladder tumors was increased in topiramate-treatment mice in the 21-month study (statistical significance was reached for HD males and females). This was largely due to the increased occurrence of a tumor that was initially diagnosed as a leiomyosarcoma but that was considered unusual from a histomorphological standpoint. These lesions were described as variable-sized nodular proliferations of large pleomorphic cells in the submucosa and muscular wall of the bladder. None of the nodules had metastasized, and only one was visible grossly. Although its incidence was increased in treated mice, this bladder lesion was also seen in two controls. When they were examined by outside experts (hired by the sponsor), there was no agreement on whether or not the lesions in question were really leiomyosarcomas or even neoplastic, but all three consultants indicated that they were unique to mice and of little or no clinical significance. One pathologist determined that only two of the lesions were malignancies, one control and one HD. Spontaneous and induced urinary bladder tumors in experimental animals and humans are usually transitional cell carcinomas; smooth muscle tumors are uncommon, and the type seen in the topiramate study has apparently only been described in mice, particularly in Swiss-derived strains such as the one used in this study (Jabobs et al., 1976). Chandra and Frith (1991) reported a low spontaneous incidence (0.375%) of bladder tumors with the same morphological characteristics as those in the present study, in CD-1 mice. Because the bladder lesions in the topiramate study were always accompanied by inflammatory changes and often by urothelial hyperplasia, one consultant concluded that they represented a proliferative response to chronic irritation. Calculi were found in the bladders of two HD females with tumors. An association between the presence of calculi (or various other physical and chemical irritants) in the bladder lumen and the development of bladder epithelial hyperplasia and neoplasia is well established in rodents. In fact, tumors like those classified as leiomyosarcomas in this study were first described in mice following surgical implantation of wax pellets in the urinary bladder (Bonser and Jull, 1956). Urothelial hyperplasia and bladder tumors seen in mice treated with 4-ethylsulfonylnaphthalene-1-sulfonamide (ENS) have been linked to urine alkalinization and calculus formation resulting from inhibition of CA by ENS. Neutralizing the urine with ammonium chloride reportedly prevented urolithiasis and the development of hyperplasia and tumors in ENS-treated mice. Urothelial hyperplasia has been seen after administration of other CA inhibitors such as acetazolamide to rodents (but not rabbits, dogs, or monkeys) and may in some cases progress to neoplasia. These bladder effects were also thought to be secondary to the changes in urine composition produced by CA inhibition. Since topiramate is a CA inhibitor and has been shown to elevate urine pH and induce calculus formation and urothelial hyperplasia in rodents, a similar pathogenesis of bladder neoplasia in topiramate-treated mice seems likely. Although most of the bladder tumors increased by topiramate treatment were of a type regarded as species specific, single incidences of hemangiosarcoma, transitional cell papilloma, and transitional cell carcinoma in HD female mice indicate that the bladder proliferative changes produced by topiramate may sometimes (but rarely) progress to tumors with human relevance. No statistically significant increase in tumors was seen in the rat carcinogenicity study, but transitional cell papillomas found in the urinary bladders of 1 MD and 2 HD rats and single incidences of urinary bladder leiomyosarcoma (MD) and renal tubular carcinoma (HD) could have been related to the alterations in urinary physiology produced by topiramate, as described above. Glandular mucosal hyperplasia of the stomach was seen in both the mouse and the rat carcinogenicity studies, but there was no evidence of progression to neoplasia. The stomach tumors seen in these studies primarily involved the forestomach. Genotoxicity results for topiramate in a full battery of tests were negative, supporting an epigenetic mechanism for the induction of bladder tumors by topiramate. In view of the low incidence of bladder tumors with human relevance associated with topiramate administration to mice and their probable etiology, the findings do not indicate that the drug presents a significant carcinogenic risk to patients.

Developmental

Topiramate is clearly teratogenic in mice, rats, and rabbits. In mice, malformations were increased at all doses studied, indicating that the threshold dose for malformations is as low as 20 mg/kg in this species. The malformations consisted primarily of craniofacial defects, including cleft palate, ablepharon, and exencephaly. Although these are among the most common sporadic malformations in mice, a variety of known teratogens can increase their incidences. Limb reduction defects were found in a single fetuses at the LD only, but CD-1 mice are considered resistant to the induction of limb reduction defects by acetazolamide. Evidence of maternal toxicity (decreased weight gain) was seen only at the HD in the mouse study, indicating a selective effect on the embryo. In rats, limb malformations with low spontaneous incidences (ectrodactyly, polydactyly, micromelia, and limb agenesis) were observed with a combined litter incidence of approximately 27% (7% of fetuses) at the HD (500 mg/kg). All but the single case of polydactyly appeared to be typical of those produced in rodents by CA inhibitors, although characterization was incomplete. The same kind of limb malformations were also found in single fetuses in the MD (100 mg/kg) and LD (20 mg/kg) groups, but in none of the control group fetuses. Both lower dose fetuses apparently also exhibited features of a spontaneously occurring genetic abnormality referred to as "short thick body syndrome," leading the sponsor to question the causal role of topiramate in these cases. However, this syndrome, which is characterized by shortening of the torso and vertebral fusion and disorganization, does not normally include limb effects. It is possible, though, that the genetic backgrounds of these fetuses increased their sensitivity to topiramate-induced limb defects. Skeletal variations were increased at the MD and HD and fetal BW was decreased at all doses in this study, suggesting that the threshold for embryotoxicity was as low as 20 mg/kg. Decreased maternal weight gain was also seen at all doses, but clinical signs of maternal toxicity were noted only at the HD. In a second rat teratology study, limb reduction defects (right-sided ectrodactyly) were only seen at the highest dose (400 mg/kg); however, the next highest dose was 30 mg/kg. Visceral anomalies were dose-dependently increased in this study due to increased incidences of thymic remnant and hydronephrosis in treatment group fetuses. Both defects occur spontaneously with average frequencies of 5-10% in the strain of rats used in this study, so their classification as malformations is questionable; they are usually considered variations, and their increased incidence probably reflects treatment-related developmental delay. Like increases in skeletal variations, treatment-related increases in the frequencies of these visceral anomalies can be an indication that the embryotoxic range of dosage is being approached. They would not be expected to have any long-term consequences, however. Embryotoxicity was evident in the persistent growth deficits seen in offspring exposed to the two highest doses in this study, while maternal toxicity was observed only at the HD. Based on the findings as a whole, 2.5 mg/kg was the highest no adverse effect dose for development toxicity in rats. It is difficult to set a threshold for malformations, but it is probably between 20 and 100 mg/kg. In the rabbit teratology studies, malformation frequencies (vertebral and rib defects, blood vessel anomalies) were increased at a dose of 120 mg/kg, and other signs of embryotoxicity (increased intrauterine death, growth retardation) were seen at 35 mg/kg or greater; 10 mg/kg was the no effect dose for developmental toxicity.

The spectrum of malformations seen with topiramate is similar to that found in teratology studies with other CA inhibitors. These agents produce a characteristic limb reduction defect when administered to pregnant rodents during a specific period of organogenesis, affecting the right side preferentially. A high incidence of right forelimb postaxial ectrodactyly has been reported after administration of single doses of acetazolamide (250-1500 mg/kg) to mice or rats during the critical period for limb development (E9-11). This malformation is thought to result from selective perturbation of the inductive process responsible for the genesis of the apical ectodermal ridge, probably secondary to a transient acidosis. Postaxial forelimb ectrodactyly predominantly of the right side has also been associated with exposure to other agents such as ethanol, cadmium, or CO2 that are thought to induce an acidotic embryonic environment. With some exposures, bilateral limb defects, hindlimb effects, and more extensive limb reductions (micromelia, amelia) may be produced. Other teratogenic effects have also been seen after administration of acetazolamide to pregnant animals. For example, a variety of malformations including fused ribs and vertebrae, gastroschisis, tail defects, cleft palate as well as ectrodactyly were observed when acetazolamide (300

mg/kg po) was administered to pregnant CD-1 mice on days 6-15 of gestation. This dose also produced a very high rate of resorption An increase in vertebral and costal skeletal malformations has been reported after administration of acetazolamide to rabbits throughout organogenesis, at oral doses as low as 50 mg/kg. Limb reduction defects have not been observed experimentally following acetazolamide administration to non-rodents, including primates, and retrospective surveys of CA inhibitor use during pregnancy have not demonstrated an increased risk of birth defects associated with these agent in humans. (In this regard, it would be useful to examine the teratogenicity of topiramate in monkeys). Species and strain differences in sensitivity may reflect differences in metabolism, carbonic anhydrase activity, or compensatory ability (eg, Na^+/H^+ antiporter functional capacity). Acetazolamide produced more pronounced decreases in maternal plasma, amniotic fluid, embryonic plasma, and embryonic intracellular pH values in the teratogenically sensitive C57 strain than in the resistant SWV strain of inbred mice (Scott et al., '90). Dose-limiting metabolic acidosis and rapid tolerance development may also be responsible for the absence of teratogenicity in humans treated with CA inhibitors. Although the teratogenic response to topiramate appears to be consistent with CA inhibition, it cannot be assumed that the developmental effects are mediated via this mechanism, since topiramate is thought to have multiple mechanisms of actions. While induction of limb reduction defects by acetazolamide or other CA inhibitors is generally associated with severe and prolonged maternal metabolic acidosis, it was not clear that such effects were seen in topiramate-treated dams. The sponsor should measure the pH of maternal blood (and blood gases) and possibly embryonic pH following teratogenic doses of topiramate in order to support their contention that inhibition of CA is the likely means by which topiramate initiates teratogenesis. The teratogenic potency of topiramate relative to other CA inhibitors is difficult to assess, but based on the available animal studies topiramate seems to be a less potent teratogen than acetazolamide. However, differences in CA isozyme inhibitory profiles could make human effects hard to predict.

Conclusions

Taken as a whole, the toxicology findings appear to primarily represent consequences of CA inhibition and, with the exception of developmental effects, do not indicate that the potential for serious toxicity with topiramate is very great. NOAELS were generally below the maximum clinical dose, so there is no safety margin for many of the effects observed in terms of dose. However, there were marked species differences in the CA inhibitory activity of topiramate, and topiramate inhibited the rat form of one CA isozyme (CA II) about 100 times more potently than the human form. Based on the animal data, the clinical side effects of topiramate might be expected to resemble those associated with acetazolamide. Although the *in vitro* carbonic anhydrase inhibitory potency of acetazolamide ranged from several orders of magnitude to <10-fold greater than that of topiramate, depending on the species and CA isozyme evaluated and the assay system employed, acetazolamide and topiramate were approximately equipotent with respect to anti-MES activity and exhibited comparable gi and renal activities in rats *in vivo*. The acetazolamide dose range in adults with epilepsy (10 to 20 mg/kg/day) includes the anticipated maximum human daily dose of topiramate (800 mg). Serious toxicity with acetazolamide and other carbonic anhydrase inhibitors is rare, and acetazolamide is considered one of the least toxic AEDs. Paresthesia, hearing dysfunction or tinnitus, gi disturbances, metabolic acidosis, decreased libido, and instances of drowsiness and confusion have been reported with therapeutic use of acetazolamide. Calculus formation and ureteral colic are fairly common. Sulfonamide-type hypersensitivity reactions have only infrequently been reported. Aplastic anemia has been reported with an incidence as high as 1 in 18,000 patient-years for all acetazolamide users, but has not been seen in epilepsy patients. Several pharmacologic properties in addition to CA inhibition (Na^+ channel blockade, potentiation of GABA receptor responses, inhibition of kainate/AMPA receptor activity) have been identified that may account for or contribute to the anticonvulsant activity of topiramate, however, and these could also contribute to the drug's toxicity. It is possible that additional central actions of topiramate are involved in the CNS toxicity reported in some of the animal studies. Cognitive dysfunction has been described in clinical trials which is reportedly more serious than any CNS side effects associated with acetazolamide, and such effects may not be apparent in preclinical studies without specific neurotoxicological testing.

Labeling

Mechanism of action

The carbonic anhydrase inhibitory potency of acetazolamide ranged from several orders of magnitude to <10-fold greater than that of topiramate, depending on the species and CA isozyme evaluated and the assay system employed. Therefore, in the last sentence, "much weaker " should be changed to "generally weaker."

Carcinogenesis, Mutagenesis, Impairment of Fertility

An increased number of urinary bladder tumors was observed in mice given topiramate in the diet for 21 months (20, 75, or 300 mg/kg/day). The elevated bladder tumor incidence (statistically significant in high dose group males and females) was largely due to the increased occurrence of a smooth muscle tumor considered histomorphologically unique to mice; therefore, the relevance of this finding to human carcinogenic risk is uncertain. Plasma topiramate exposures (based on AUC) in mice receiving the high dose of 300 mg/kg were approximately 0.3 - 0.5 times the highes exposures measured in patients treated with topiramate (at a daily dose of 800 mg). No evidence of carcinogenicity was seen in rats following oral administration of topiramate for 2 years at doses up to 120 mg/kg/day, which is approximately 1.5 times a human daily dose of 800 mg on a mg/m2 basis.

Topiramate did not demonstrate mutagenic or genotoxic potential when tested in a battery of *in vitro* and *in vivo* assays. Topiramate was negative for mutagenicity in bacteria and in mouse lymphoma cells *in vitro*; did not increase unscheduled DNA synthesis in rat hepatocytes *in vitro*; and did not significantly increase chromosomal aberrations in human lymphocytes *in vitro* or in rat bone marrow *in vivo*.

No adverse effects on male or female fertility were observed in rats at doses up to 100 mg/kg, or approximately 1 times a human daily dose of 800 mg on a mg/m2 basis.

Pregnancy

Pregnancy Category C: Topiramate has been shown to be teratogenic in mice, rats, and rabbits. When oral doses of 20, 100, or 500 mg/kg were administered to pregnant mice during the period of organogenesis, the frequency of fetal malformations (primarily craniofacial defects) was increased in all drug-treated groups. Fetal weight and skeletal ossification were reduced at the high dose in conjunction with decreased maternal weight gain. The low dose in this study is approximately 0.1 times a human daily dose of 800 mg on a mg/m2 basis. When pregnant rats received oral doses of 20, 100 or 500 mg/kg throughout organogenesis, an increased incidence of fetal limb malformations (ectrodactyly, micromelia, amelia; predominantly right-sided) was observed in litters exposed to the high dose. This dose, which produced clinical signs of maternal toxicity, represents approximately 6 times a human daily dose of 800 mg on a mg/m2 basis. Dosage-related reductions in fetal weight and maternal weight gain were noted in all drug-treated groups. The low dose is approximately 0.2 times a human daily dose of 800 mg on a mg/m2 basis. In a second rat teratology study (0.2, 2.5, 30 or 400 mg/kg during organogenesis), the incidence of right forelimb ectrodactyly was increased in the offspring of dams treated with an oral dose of 400 mg/kg, or approximately 5 times a human daily dose of 800 mg on a mg/m2 basis. This dose was also maternally toxic. Doses exceeding 2.5 mg/kg (>0.03 times a human daily dose of 800 mg on a mg/m2 basis) produced evidence of embryotoxicity (increased incidence of anatomical variants, retarded growth). In a rabbit teratology study (10, 35, or 120 mg/kg during organogenesis), fetal malformations (vertebral and rib defects, blood vessel anomalies) were

increased after treatment of pregnant animals with an oral dose of 120 mg/kg or 2.4 times a human daily dose of 800 mg on a mg/m2 basis. Doses exceeding 10 mg/kg (>0.2 times a human daily dose of 800 mg on a mg/m2 basis) were embryotoxic (intrauterine death, reduced fetal weight), and maternal toxicity occurred over the same dose range. When female rats were treated with 0.2, 4, 20, or 100 mg/kg during the last third of gestation and throughout the lactation period, offspring weights were decreased at birth and during lactation in groups receiving doses greater than 4 mg/kg (>0.05 times a human daily dose of 800 mg on a mg/m2 basis). The high dose produced maternal toxicity during the gestational treatment period.

One of the pharmacological actions of topiramate is inhibition of carbonic anhydrase. Malformations similar to those produced by topiramate (limb reduction defects in rodents, axial and costal skeletal defects in rabbits) have been found in teratology studies with other carbonic anhydrase inhibitors. Although it is well established that carbonic anhydrase inhibitors can induce these abnormalities in rodents and rabbits, epidemiological studies have not identified an increased risk of birth defects associated with the use of these agents during human pregnancy. However, it cannot be assumed that all of the developmental effects of topiramate are mediated via this mechanism or that human responses to topiramate can be predicted from the experience with other carbonic anhydrase inhibitors. There are no studies of topiramate in pregnant women.

XI. RECOMMENDATIONS

The NDA is approvable with respect to the pharmacology/toxicology portion. However, additional characterization of the teratogenic action of topiramate is needed. The sponsor should measure maternal plasma pH and/or CO2 following teratogenic doses of topiramate in order to support their contention that inhibition of CA is the likely means by which topiramate initiates teratogenesis. This can be done in Phase 4. Recommendations concerning the proposed labeling are made in the Evaluation section of the review.

cc:
NDA (20-505)
Div File
HFD-120/GFitzgerald/EFisher/RPitts

E. Fisher, Ph.D.

GFF 11/9/95
Team Leader's comment:
It is not necessary for the sponsor to conduct the recommended studies to characterize the teratogenic activity of topiramate unless they wish to attribute the observed teratogenicity in labeling to a specific mechanism.
GFF

References

Bonser GM, and JW Jull (1956) The histological changes in the mouse bladder following surgical implantation of paraffin wax pellets containing various chemicals. J Pathol Bacteriol, 72:489-495.

Chandra M, and CH Frith(1991) Spontaneously occurring leiomyosarcomas of the mouse urinary bladder. Toxicol Pathol, 19:164-167.

Jacobs JB, Cohen SM, Arai M, Friedell GH, Bulay O, and HK Urman (1976) Chemically induced smooth muscle tumors of the mouse urinary bladder. Cancer Res 36:2396-2398.

Scott WJ, Duggan CA, Schreiner CM, and MD Collins (1990) Reduction of embryonic intracellular pH: a potential mechanism of acetazolamide-induced limb malformations. Tox Appl Pharmacol, 103:238-254.

END

BT

J.H.M. Research & Development, Inc., 5776 Second Street, N.E., Washington, D.C. 20011