processed, and analyzed for drug-related materials. Separation and identification of metabolites were accomplished using , system. One polar metabolite, M4, was methylated using : for analysis.

Results: in urine (3/sex), a total of 11 peaks were detected in both male and female rats. The metabolic profile in urine was similar between sexes, although there were quantitative differences. The following urinary metabolites were identified: Peak II (M1), Peak III (M2), Peak IV (M3), Peak V (M4), Peak VI (M5, M6), Peak VII (M7): 6-chloro-5-[2-{4-(2-sulfinamido)benzoyl}piperazinyl]ethyl-oxindole, Peak VIII (M8): CP-88,059 sulfone, Peak IX (M9), Peak X (M10), Peak XI (M13).

Parent compound (M13) accounted for 0.05-1.07 and 4.64-5.64% of urinary radioactivity in males and females, respectively. Of the metabolites detected in males, M4 was the major metabolite, accounting for ≈42% of urinary radioactivity. M5 + M6 and M2 accounted for 24.41-32.28 and 22.88% of urinary radioactivity, respectively. In females, M4 was the major metabolite, accounting for 28.86% of urinary radioactivity. M9, M5 + M6, and M10 were also fairly abundant, accounting for 17.22-21.84, 19.93-32.09, and 8.02-10.21% of urinary radioactivity, respectively. M1, M2, M3, and M4 represented cleavage products. Total recovery of radioactivity was ≈85-95%.

In bile (3/sex), a total of 11 peaks were detected in both male and female rats. Metabolites were identified in bile samples from 1/sex. Five major metabolites were identified (accounting for 66% of total radioactivity), with four of these also being detected in urine. The metabolites were identified as follows: Peak IV (M6), Peak V (M7), Peak IX (M9), Peak X (M12), and Peak XI (M13). Six metabolites were detected, but not identified (Peaks I-III, VI-VIII).

Parent compound accounted for a greater portion of biliary radioactivity in females than in males (15.26-17.10 and 6.07-4.96, respectively). Of the metabolites detected, M6, M7, and M9 were major metabolites in males, accounting for 16.09-22.40, 22.11-28.24, and 11.73-16.79% of biliary radioactivity, respectively. In females, M9 was the major biliary metabolite, accounting for 21.24-21.50% of biliary radioactivity. M6 and M7 were also fairly abundant, accounting for 11.51-15.15 and 12.31-14.86% of biliary radioactivity, respectively. Total recovery of radioactivity was ≈92-99%. All identified metabolites appeared to be intact molecules.

In feces (3/sex), the parent compound accounted for a majority of total fecal radioactivity (91-95%). Three minor metabolites were detected, accounting for ≈7% of fecal radioactivity. Fecal samples from 1/sex were used to identify the metabolites. All three minor metabolites resulted from oxidation at the BITP moiety: Peak I (M9), Peak II (M10), Peak III (M11): CP-88,059 piperazinyl-N-oxide, Peak IV (M13). M9, M10, and M11 accounted for 0.34-0.93%, 2.14-4.63, and 2.10-2.97% of fecal radioactivity, respectively. Total recovery of radioactivity was >99%.

The data were summarized for males and females separately in the sponsor's Tables 4 and 5 (below). Based on the structures of the identified metabolites, the sponsor summarized the metabolism of CP-88,059 in rat as follows:

Major routes:

(1) "...N-dealkylation of ethyl side chain attached to the piperazinyl nitrogen..."

(2) "...oxidation at [the] sulfur resulting in the formation of sulfoxide and sulfone..."

(3) "...oxidation on the benzisothiazole moiety (other than [the] sulfur)..."

(4) "...hydroxylation on the 3-position and subsequent oxidation of the benzisothiazole moiety..."

Minor routes: "...N-oxidation at the piperazine and hydrolysis of the oxindole moiety..."

Table 4. Percentages of metabolites of CP-88,059 in male rats.

Metabolite	% of Dose[a]			
	Urine[b]	Bile[c]	Feces[d]	Total
M1[e]	1.13	ND	ND	1.13
M2[e]	5.14	ND	ND	5.14
M3[e]	2.08	ND	ND	2.08
M4[f]	9.24	ND	ND	9.24
M5+M6[g]	7.25	3.00	ND	10.25
M7	1.05	3.92	ND	4.97
M8	0.27	ND	ND	0.27
M9	1.54	2.22	0.40	4.16
M10	0.76	ND	1.70	2.46
M11	ND	ND	1.35	1.35
M12	ND	0.26	ND	0.26
M13	0.12	0.85	59.36	60.33

a; average of ^{3}H and ^{14}C
b; based on 22.46(^{3}H) and 21.89% (^{14}C) recovery
c; based on 16.2(^{3}H) and 14.68% (^{14}C) recovery
d; based on 63.63% (^{3}H) and 62.21% (^{14}C) recovery
e; only ^{3}H label
f; only ^{14}C label
g; not separated on

Table 5. Percentages of metabolites of CP-88,059 in female rats.

Metabolite	% of Dose[a]			
	Urine[b]	Bile[c]	Feces[d]	Total
M1[e]	0.76	ND	ND	0.76
M2[e]	0.98	ND	ND	0.98
M3[e]	0.21	ND	ND	0.21
M4[f]	5.38	ND	ND	5.38
M5+M6[g]	5.00	2.75	ND	7.75
M7	1.46	2.80	ND	4.26
M8	1.14	ND	ND	1.14
M9	3.75	4.39	0.43	8.57
M10	1.75	ND	2.16	3.91
M11	ND	ND	1.50	1.50
M12	ND	0.99	ND	0.99
M13	0.98	3.33	46.56	50.87

a: average of ^{3}H and ^{14}C
b: based on 22.46% (^{3}H) and 21.89% (^{14}C) recovery
c: based on 16.2% (^{3}H) and 14.68% (^{14}C) recovery
d; based on 63.63% (^{3}H) and 62.21% (^{14}C) recovery
e: only ^{3}H label
f; only ^{14}C label
g; not separated on f

The proposed metabolic pathways for CP=88,059, based on the metabolic data in urine, bile, and feces in rats is summarized in the following sponsor's Figure 65:

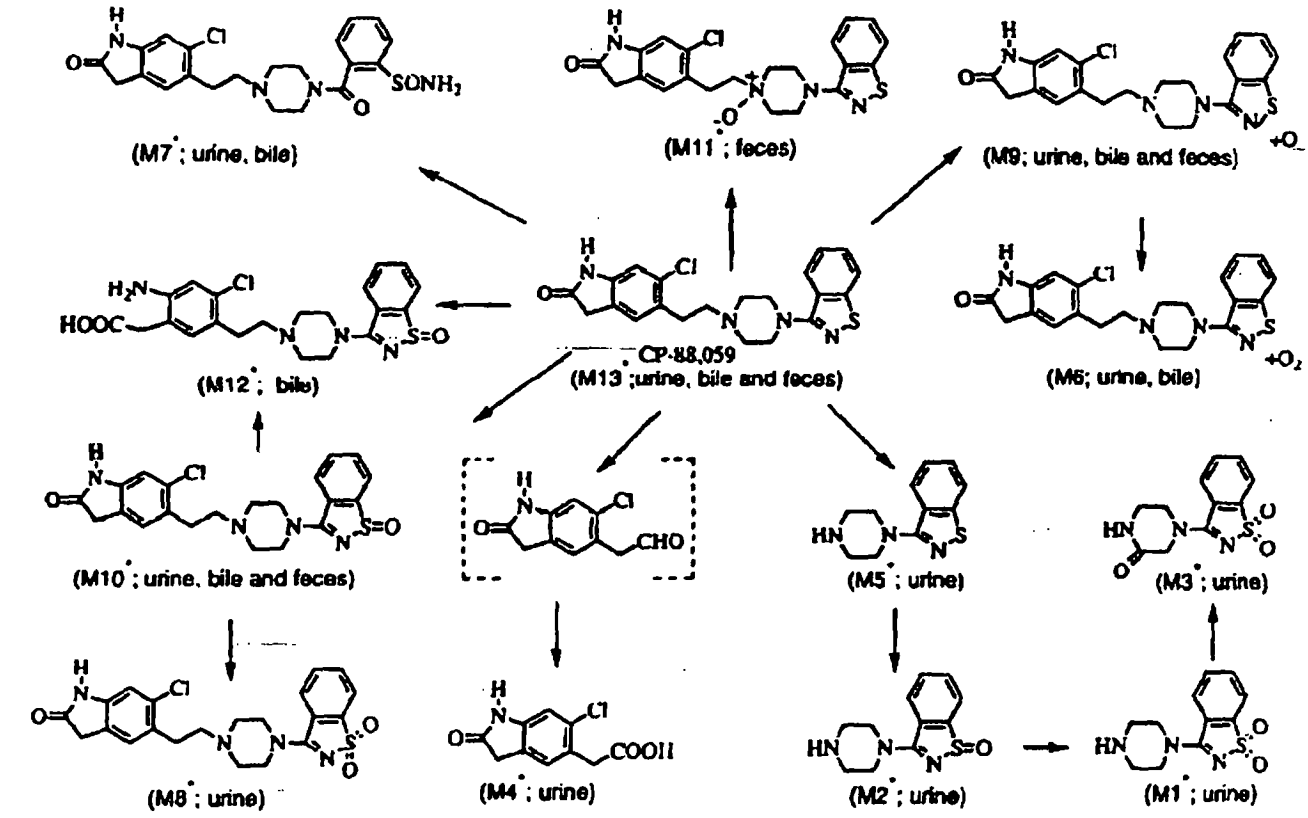

Figure 65. Proposed routes for the biotransformation of CP-88,059

Dog

1. **Pharmacokinetics and oral bioavailability of CP-88,059 in Beagle dogs** (Report No. DM-93-128-4)

Methods: CP-88,059 (lot no. not specified) was administered as single p.o. (2.0 mg/kg) and i.v. (0.5 mg/kg) doses to 4 male Beagle dogs. Each dog received both a p.o. and an i.v. dose. Blood samples were collected at 0.25, 0.5, 1, 1.5, 2, 3, 5, and 6 hr after the i.v. dose and at 1, 2, 3, 4, 6, 8, and 12 hr after the p.o. dose. Plasma drug levels were quantitated by following organic extraction of plasma samples. The LLOQ was ng/mL.

Results: the data are summarized in the following sponsor's table:

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TABLE 3
PHARMACOKINETIC SUMMARY OF CP-88,059 IN MALE DOGS

IV DOSE

DOG #	Dose (mg/kg)	AUC (0-inf) (ng-hr/ml)	AUMC (0-inf) (ng-hr*hr/ml)	Cl (ml/min-kg)	Vdss (L/kg)	Kel (1/hr)	T1/2 (hr)
33664	0.5	559	1608	14.9	2.6	0.267	2.4
34239	0.5	589	1537	14.1	2.2	0.310	2.2
34240	0.5	512	1329	16.3	2.5	0.286	2.4
34241	0.5	479	1042	17.4	2.3	0.324	2.1
Mean		635	1377	15.7	2.4	0.302	2.3
S.D.		49	285	1.4	0.2	0.019	
%C.V.		9	18	9.2	7.4	6.1	

PO DOSE

DOG #	Dose (mg/kg)	AUC (0-inf) (ng-hr/ml)	Bioavailability (%)	Plasma Conc (ng/ml)	Tmax (hr)	Kel (1/hr)	T1/2 (hr)
33664	2	542	24	70	2	0.202	3.4
34239	2	750	32	168	1	0.239	2.9
34240	2	867	42	135	2	0.289	2.4
34241	2	839	44	182	1	0.275	2.5
Mean		752	36	139	1.5	0.251	2.8
S.D.		147	9	50	0.58	0.039	
%C.V.		20	25	36	39	15.5	

The V_d estimate indicates fairly limited tissue distribution of the parent compound. The Cl estimate, 15.7 mL/min/kg is well below the hepatic blood flow (estimated to be 25-43 mL/min/kg in this species).

2. **Radiolabel excretion, circulating radioactivity and unchanged drug in bile duct-cannulated Beagle dogs after oral administration of [^{3}H]- and [^{14}C]-labeled CP-88,059** (Study no. DM-94-128-16)

Methods: bile-duct cannulated Beagle dogs (2/sex) were dosed with a mixture of [^{3}H]- and [^{14}C]-labeled CP-88,059 (unlabeled lot no. 20480-224-1) as a single oral dose (5 mg/kg; vehicle was 0.3% methylcellulose). Urine, bile, and fecal samples were collected as follows: (1) urine and bile samples were collected for 168 hr (7 days) postdosing at 0.6-, 24-48, 48-72, 72-96, 96-120, 120-144, and 144-168 hr postdosing and (2) fecal samples were collected at 24-hr intervals up to 168 hr postdosing. Blood samples were collected at 0.5, 1, 2, 4, 6, 8, 12, and 24 hr postdosing. Sample radioactivity was quantitated using following processing. Plasma levels of parent compound were quantitated using

Results: the data are summarized in the following table:

SEX	URINE	FECES	BILE	PLASMA	
	% of Dose Radioactivity			C_{max} (ng/mL)	$AUC_{(0-T)}$ (ng•hr/mL)
M	5.9-6.0	86.3-87.5	8.2-9.3	132 (28-31)#	428 (12-16)
F	15.3-15.4	79.7-80.1	8.6-9.0	277 (34)	2008 (19-21)

#(% of total radioactivity)

Total recovery of radioactivity was ≈100% in both males and females. The % of radioactivity secreted in bile was similar in males and females, whereas that excreted in urine and feces differed depending upon the sex. The small amount of biliary radioactivity would suggest that the majority of fecal radioactivity is accounted for by either unabsorbed material or by GI secretion.

3. **Identification of metabolites in plasma of Beagle dogs after oral administration of [^{3}H]- and [^{14}C]-labeled CP-88,059-1** (Study no. DM-95-128-17).

Methods: radiolabeled CP-88,059-1 (unlabeled lot no. 20480-224-1) was administered to Beagle dogs (2/sex) at a single oral dose of 5 mg/kg (vehicle: 0.5% methyl cellulose). Blood samples were collected at 0, 0.5, 1, 2, 4, 6, 8, and 12 hr postdosing. Samples collected at 1, 2, and 4 hr were pooled for identification of metabolites. Metabolites were separated and identified using

Results: 8 peaks were detected in plasma of both male and female dog (1,4 hr samples): Peak I (M1), Peak II (M2), Peak III (M4), Peak IV (M5), Peak IV (M6), Peak V (M7): 6-chloro-5-[2-{4-(2-sulfinamido)benzoyl}piperazinyl]ethyl-oxindole, Peak VI (M8): CP-88,059 sulfone, Peak VII (M9), Peak VII (M10), Peak VIII (M13).

The data are summarized in the following sponsor's Tables 1 and 2. The metabolic profile was fairly similar between isotopes, indicating the metabolites represented intact molecules. The parent compound was the single compound that accounted for the majority of plasma radioactivity; although M9 and M10 together accounted for the majority of plasma radioactivity. M8 was a major metabolite (i.e., accounted for ≥10% of plasma radioactivity) in males; in females, this metabolite was slightly less abundant. Together the 9 metabolites and the parent compound accounted 70-83% of total plasma radioactivity.

Table 1. Percentage of circulating metabolites of CP-88,059 in male beagle dogs after oral administration of [^{3}H]-and [^{14}C] CP-88,059.

Metabolite (#)	Retention Time (min)	% of Radioactivity (1 hr)		% of Radioactivity (4 hr)	
		3H	14C	3H	14C
M1	7.9	1.2	1.1	1.9	1.6
M2	10.7	3.5	1.7	4.4	1.3
M4	22.6	2.1	2.9	4.7	2.7
M5 & M6	33.2	3.9	3.3	3.1	3.3
M7	35.7	6.1	5.6	6.7	7.6
M8	38.8	10.4	11.5	18.1	21.2
M9 & M10	40.5	27.6	31.3	24.7	28.6
M13	48.4	18.4	20.4	14.7	16.5
Total		73.3	77.7	78.2	82.7

Table 2. Percentage of circulating metabolites of CP-88,059 in female beagle dogs after oral administration of [^{3}H]-and [^{14}C] CP-88,059.

Metabolite (#)	Retention Time (min)	% Radioactivity (1 hr)		% Radioactivity (4 hr)	
		3H	14C	3H	14C
M1	7.9	1.6	1.3	1.8	0.6
M2	10.7	2.6	1.4	3.1	0.8
M4	22.6	1.6	5.0	1.7	2.6
M5 & M6	33.2	4.7	3.8	3.9	3.8
M7	35.7	4.3	4.1	6.6	6.8
M8	38.8	7.5	7.7	13.4	14.4
M9 & M10	40.5	26.4	26.2	23.5	25.4
M13	48.4	22.1	22.6	18.7	20.2
Total		70.8	72.1	72.6	74.8

The metabolic profile of CP-88,059 in dogs is summarized in the following sponsor's figure:

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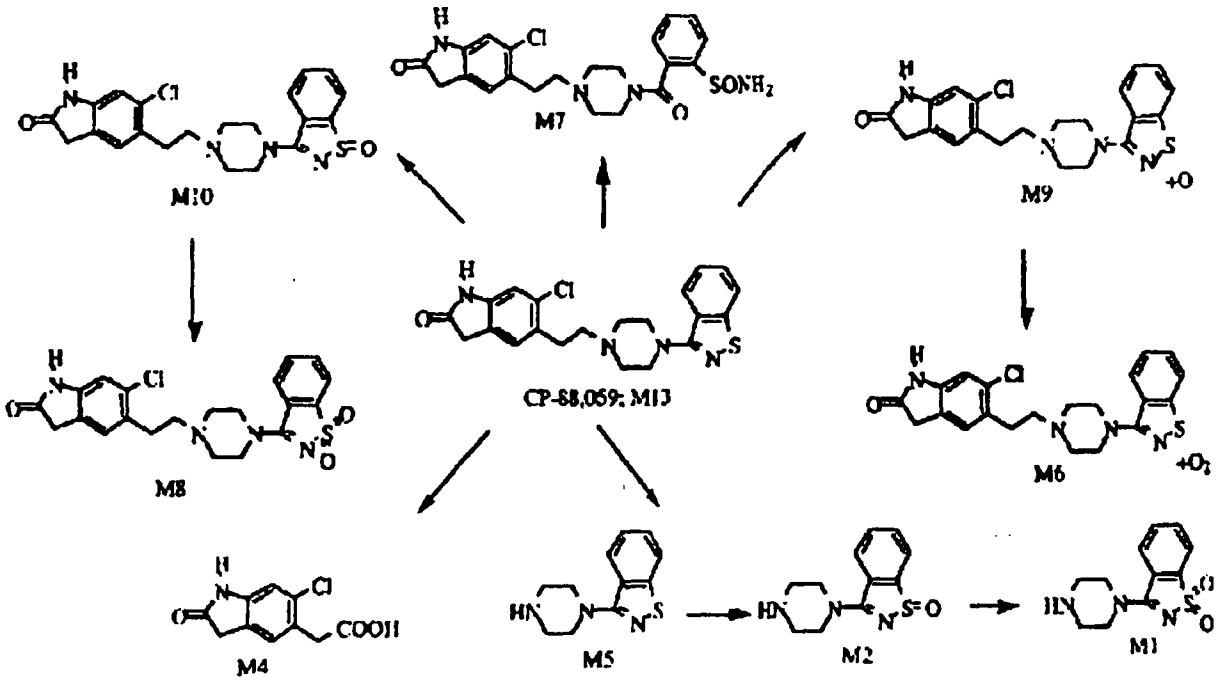


Figure 13. Proposed metabolites of CP-88,059 in dog plasma.

4. **Identification of metabolites in urine, bile and feces of Beagle dogs after oral administration of [^{3}H]- and [^{14}C]-labeled CP-88,059-1** (Study #DM-95-128-26).

Methods: radiolabeled CP-88,059 (unlabeled lot #20480-224-1) was administered to bile-cannulated Beagle dogs (2/sex) as a single oral dose of 5 mg/kg (vehicle: 0.5% methyl cellulose). Urine, bile, and fecal samples were collected (time of sampling was not specified). Metabolites were separated and identified using

Results: urinary metabolites were determined on pooled samples (0-24 hrs postdosing). A total of 15 peaks were detected and were identified as follows: Peak 1 (M1), Peak II (M2), Peak III (M4), Peak IV (M14): glucuronide conjugate of 5-(2-hydroxyethyl)-6-chlorooxindole, Peak V (M4A): 5-(2-piperazinylethyl)-6-chlorooxindole, Peak VI (M15): dihydroxy-CP88,059 sulfone, Peak VII (M16): sulfate conjugate of 3'-hydroxy-CP-88,059-sulfoxide, Peak VIII (M17): hydroxy-sulfone, Peak IX (M5), Peak IV (M6), Peak IX (M7), Peak X (M8), Peak XII (M9), Peak XII (M10), Peak XIII (M13).

M1, a cleaved molecule, was the major urinary metabolite in males and females. The identified metabolites and parent compound accounted for ≈80-97% of urinary radioactivity.

Biliary metabolites were determined on pooled samples (0-6 hrs postdosing). A total of 8 major peaks were detected. These were identified as follows: Peak I (M16), Peak II (M6), Peak II (M7), Peak III (M7A): intact molecule with addition of 2 oxygen atoms at the benzisothiazole moiety, Peak IV (M18): dihydroxy-sulfoxide, Peak V (M8), Peak VI (M10), Peak VII (M9), Peak VIII (M13).

M9, M10, and the parent compound accounted for the majority of biliary radioactivity; in

females. Interestingly, M9 was clearly the major drug-related compound in the bile of 1 F (34-38%), but not in the other (8-10%). In the second F, the parent compound was the major drug-related compound (25-29% of biliary radioactivity) A similar finding was noted in males, although the difference between males was not quite as notable (≈18 vs 25-30%). In the 2 males, the parent compound accounted for a similar portion of biliary radioactivity (15-16 and ≈17%). The identified metabolites and the parent compound accounted for 76-95% of total biliary radioactivity.

Fecal metabolites were determined in samples collected at 0-24 hr postdosing. The majority of radioactivity was associated with the parent compound(80-95% of total fecal radioactivity; 5 minor metabolites were detected. These were identified as follows: Peak I (M4A): 5-(2-piperazinylethyl)-6-chlorooxindole, Peak II (M8), Peak III (M9), Peak III (M10), Peak IV (M11): CP-88,059 piperazinyl-N-oxide, Peak V (M13). M4A was detected only in male dog. The identified material accounted for 92-97% of total fecal radioactivity.

The data (expressed as % dose) are summarized in the following sponsor's Tables 6 and 7:

Table 6. Percentage of metabolites of CP-88,059 in male dogs

Metabolite #	% of Dose[a]			
	Urine[b]	Bile[c]	Feces[d]	Total
M1[e]	2.22	ND	ND	2.22
M2[e]	0.61	ND	ND	0.61
M4[f]	2.46	ND	ND	2.46
M14[f]	0.30	ND	ND	0.30
M4A[f]	0.05	ND	5.75	5.80
M15	0.02	ND	ND	0.02
M16	0.46	0.44	ND	0.90
M17	0.06	ND	ND	0.06
M5-M6-M7[g]	0.50	0.59	ND	1.09
M7A	ND	0.42	ND	0.42
M18	0.06	ND	ND	0.06
M8	0.31	0.69	1.90	2.90
M9+M10[g]	0.74	3.00	1.45	5.18
M11	ND	ND	1.16	1.16
M13	0.13	1.43	73.78	75.34

a: average of (^{3}H) and (^{14}C) labels.
b: based on 6.0% (^{3}H) and 5.9% (^{14}C) recovery.
c: based on 9.3% (^{3}H) and 8.2% (^{14}C) recovery.
d: based on 87.5% (^{3}H) and 86.3% (^{14}C) recovery.
e: only (^{3}H) label.
f: only (^{14}C) label.
g: not separated on

Table 7. Percentage of metabolites of CP-88,059 in female dogs

Metabolite #	% of Dose[a]			
	Urine[b]	Bile[c]	Feces[d]	Total
M1[e]	3.68	ND	ND	3.68
M2[e]	1.00	ND	ND	1.00
M4[f]	5.71	ND	ND	5.71
M14[f]	0.82	ND	ND	0.82
M4A[f]	0.18	ND	ND	0.18
M15	0.32	ND	ND	0.32
M16	0.23	0.30	ND	0.53
M17	0.80	ND	ND	0.80
M5-M6+M7[g]	2.68	0.34	ND	3.02
M7A	ND	0.64	ND	0.64
M18	0.39	0.55	ND	0.94
M8	0.38	1.14	0.45	1.97
M9+M10[g]	1.42	3.27	0.68	5.37
M11	ND	ND	0.37	0.37
M13	0.25	1.76	75.64	77.65

a: average of (^{3}H) and (^{14}C) labels.
b: based on 15.4% (^{3}H) and 15.3% (^{14}C) recovery.
c: based on 9.0% (^{3}H) and 8.6% (^{14}C) recovery.
d: based on 80.1% (^{3}H) and 79.7% (^{14}C) recovery.
e: only (^{3}H) label.
f: only (^{14}C) label.
g: not separated on

The proposed metabolic pathways for CP-88,059 in dog are summarized in the following sponsor's figure:

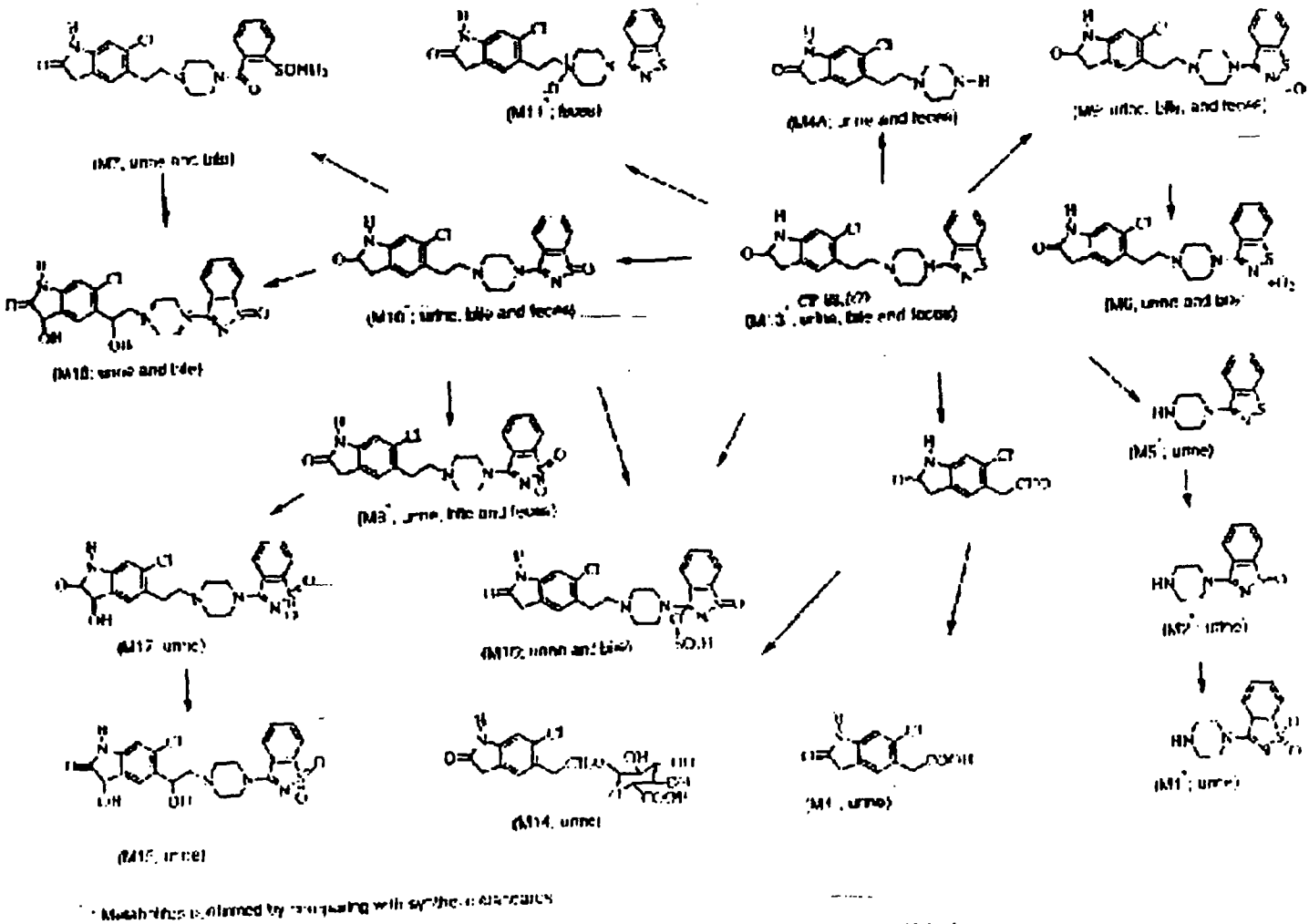


Figure 45. Proposed routes for the biotransformation of CP-88,059 in dog

M9 is thought to be formed following reduction of the parent compound and subsequent methylation by thiol S-methyl transferase.

The sponsor identified 4 major and 2 minor routes of metabolism of CO-88,059. The major route were as follows:

(1) "...N-dealkylation of ethyl side chain attached to piperazinyl nitrogen..." This route accounted for formation of M4, M5, and M14.

(2) "...oxidation at sulfur resulting in the formation of sulfoxide and sulfone..."

(3) "...oxidation on the benzisothiazole moiety (other than sulfur)..." This route accounted for formation of M6 and M9.

(4) "...hydroxylation on the 3-position and subsequent oxidation of the benzisothiazole moiety (M4A and M7) and benzylic oxidation of the oxindole moiety."

The sponsor pointed out that buspirone, tandospirone, and tiospirone (structurally related compounds) share the first two routes, and that "The formation of sulfoxide and sulfone is common with sulfur-containing drugs..." A unique finding was the oxidation on the benzisothiazole moiety other than at the sulfur. According to the sponsor, two possible sites of oxidation are (a) oxidation at the nitrogen of the benzisothiazole ring and (b) aromatic hydroxylation of the benzisothiazole moiety; "...the aromatic hydroxylation of the benzisothiazoles has not been reported in the literature."

A second "unusual" pathway was the hydroxylation of the "...3-position of the benzisothiazole moiety followed by oxidative N-debenzisothiazolation (M4A and M7). The proposed pathway for formation of these metabolites is illustrated in the following sponsor's figure:

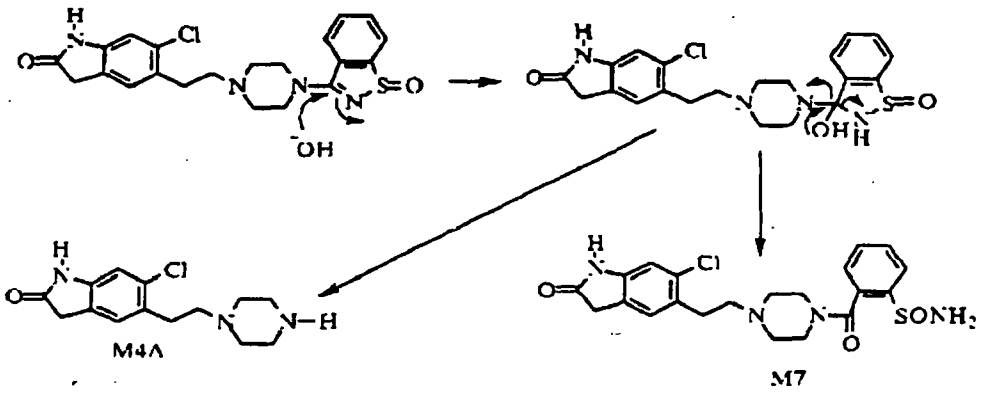


Figure 47. Proposed mechanism for the formation of metabolite M4 A and M7.

Rabbit

1. **Pharmacokinetics of CP-88,059 in the New Zealand rabbit after i.v. and i.m. administration** (Study no. DM-92-128-3).

Methods: CP-88,059 was administered to 4 grps of male rabbits according to the following scheme:

Grp 1: CP-88,059 was administered into the marginal ear vein (4 rabbits) in 40% ß-HPCD as a single dose of 2 mg/mL (0.3 mL/kg). Blood samples were collected at 0, 0.5, 1, 2, 4, 6, 8, and 24 hr postdosing.

Grp 2: 1 M received 1 mL of a 20 mg/mL aqueous suspension of CP-88,059 (≈6 mg/kg). 1 M received 1 mL of a 40 mg/mL aqueous suspension of CP-88,059 (≈13.3 mg/kg). Injections in both rabbits were i.m. Blood samples were collected at 0, 0.5, 1, 2, 4, 6, 8, 24, and 48 hr postdosing.

Grp 3: CP-88,059 (in 40% ß-HPCD, 4 males) was administered i.m. at a dose of ≈0.6 mg/kg. Blood samples were collected at 0, 0.5, 1, 2, 4, 6, 8, 24, and 48 hr postdosing.

Grp 4: CP-88,059 (aqueous suspension) or CP-88,059-1 was administered i.m. (4 males) at a single dose of ≈13.3 mg/kg. Blood samples were collected at 2, 4, 7, 24, 30, 48, 54, 72, and 78 hr postdosing.

Plasma levels of parent compound were quantitated using The limits of quantitation were g/mL to ng/mL.

Results: systemic exposure following i.m. dosing of suspensions was low and prolonged; the $AUC_{(0\text{-inf})}$ for the suspension was ≈32 ng•hr/mL and F was <0.13%. Bioavailability of the i.m. solution (at 0.6 mg/kg) was 100%. PK estimates following i.v. dosing were as follows: $AUC_{(0\text{-inf})}$ = 427 ± 56 ng•hr/mL, Cl = 23.7 ± 2.8 mL/min/kg, V_d = 2.7 ± 0.6 L/kg, and $t_{1/2}$ = 1.3 hr.

Human

1. **Radiolabel excretion, circulating radioactivity and unchanged drug in humans after oral administration of [^{3}H]- and [^{14}C]-labeled CP 88,059-1** (Study no. DM-95-128-18).

Methods: radiolabeled (dual mixture) CO-88,059 was given orally (single 20 mg dose, suspension in water) to healthy male volunteers following a "standard breakfast". Urine samples were collected at 0-4, 4-12, 12-24, 24-48, 48-72, 72-96, 96-120, 120-144, 144-168, 168-192, 192-216, 216-240 hr postdosing. Fecal samples were collected during the same 11-day period. Blood samples were collected at 0, 1, 2, 3, 4, 6, 8, 12, 16, 24, 36, 48, 72, 96, and 120 hr postdosing. Sample radioactivity was quantitated using Serum levels of parent compound were quantitated using .

Results: the PK data [means ± CV (ranges); radioactivity is expressed in ng-equiv] are summarized in the following table:

MEASUREMENT	^{3}H/^{14}C	C_{max} (ng/mL)	T_{max} (hr)	$AUC_{(0-\infty)}$ (ng•hr/mL)	$t_{1/2}$ (hr)
total radioactivity	^{3}H	94.5 ± 26 (66.4-118.7)	5.5 ± 18.2 (4-6)	876.6 ± 15.9 (805.4-1082.4)	4.52 (4.10-5.31)
	^{14}C	88.4 ± 25 (60.9-110.4)	5.5 ± 18.2 (4-6)	760.8 ± 15.6 (681.8-941.5)	3.96 (3.61-4.65)
parent compound		45.4 ± 31.5 (28.8-62.0)	3.5 ± 54.7 (2-6)	361.3 ± 11.2 (323.7-418.2)	3.53 (3.20-4.31)

Based on AUC, systemic exposure to parent compound accounted for 41-47% of total serum radioactivity.

The major route of elimination was via the feces, with fecal radioactivity accounting for 64.5-68.1% of dose radioactivity (range: %); urinary radioactivity accounted for ≈20% (range: %) of dose radioactivity. Total recovery of radioactivity was 86.4-90.6% (range: 1%). The sponsor noted that the majority of fecal and urinary radioactivity (64 and 89%, respectively) was excreted during the first 48 hr postdosing.

2. **Radiolabel excretion, circulating radioactivity and unchanged drug in humans after oral administration of [^{3}H]- and [^{14}C]-labeled CP-88,059-1** (Study no. DM-95-128-19).

Methods: a mixture of [^{3}H]- and [^{14}C]-labeled CP-88,059-1 was given orally to 4 healthy male volunteers as a single 20 mg suspension (in water) following a "standard meal". Urine was collected for 7 days postdosing (0-24, 24-48, 48-72, 72-96, 96-120, 120-144, 144-168 hr). Fecal samples were collected during the same period. Blood samples were collected at 0, 1, 2, 3, 4, 6, 8, 12, 16, 24, 36, 48, 72, 96, and 120 hr postdosing. Sample radioactivity was quantitated

using Serum levels of parent compound were quantitated using .

Results: a major route of elimination was via the feces, with fecal radioactivity accounting for 44-50% (range: '%) of dose radioactivity; urinary radioactivity accounted for ≈20% (range '%) of dose radioactivity. However, total recovery was only ≈64-71%. The sponsor suggested that sample collection may have been incomplete.

The PK data [means ± CV (ranges); radioactivity is expressed in ng-equiv] are summarized in the following table:

MEASUREMENT	$^{3}H/^{14}C$	C_{max} (ng/mL)	T_{max} (hr)	$AUC_{(0-\infty)}$ (ng•hr/mL)	$t_{1/2}$ (hr)
total radioactivity	^{3}H	75.1 ± 30.2 (50.10-96.90)	3.25 ± 30.6 (2-4)	931 ± 27.8 (649.8-1282.6)	6.73 (5.64-7.55)
	^{14}C	83.6 ± 27.2 (57.9-107.6)	3.75 ± 13.4 (3-4)	871 ± 32.1 (534.9-1213.5)	5.50 (4.52-6.38)
parent compound		53.8 ± 28.9 (33.10-67.80)	3.50 ± 16.7 (3-4)	488 ± 30.6 (299.7-637.0)	4.17 (3.45-5.04)

Based on mean AUC, the parent compound accounted for 52-56% of total systemic exposure. Serum C_{max} and AUC were notably lower in 1 of the 4 volunteers, suggesting lower absorption or incorrect dosing of this individual. However, fecal radioactivity in this individual was similar to that of the other subjects, as were serum PK estimates.

3. **Identification of metabolites in urine, feces and serum of human subjects after oral administration of [^{3}H]- and [^{14}C]-labeled CP-88,059-1** (Study no. DM-95-128-20).

Methods: a dual mixture of radiolabeled CP-88,059 was administered to 4 healthy human volunteers as a single oral dose (20 mg suspension in water) following a "standard breakfast". Detection and identification of metabolites were performed on urine (0-24 hr), fecal (24-48 hr), and serum (0-8 hr) samples using .

Results: in urine, a total of 7 radioactive peaks were detected. These were identified (using authentic standards) as follows: Peak I (M1), Peak II (M2), Peak II (M3a): glucuronide conjugate of 5-(2-carboxyethyl)-6-chlorooxindole, Peak III (M4), Peak IV (M4A): 5-(2-piperazinylethyl)-6-chlorooxindole, Peak V (M5), Peak V (M6), Peak V (M7), Peak VI (M8), Peak VI (M9), Peak VI (M10), Peak VII (M13). M1 + M2 accounted for ≈55% of urinary radioactivity; both of these metabolites represented cleaved molecules (labeled only with ^{3}H). M3A and M4 (^{14}C-labeled only) accounted for ≈27 and 35% of urinary radioactivity, respectively. Peaks V and VI accounted for ≈8 and 26-35% of urinary radioactivity, respectively. Identified metabolites and parent compound accounted for ≈97% of urinary radioactivity.

In feces, 4 metabolites and the parent compound were detected. The major fecal metabolite was identified as M9 (Peak 1). M9 accounted for ≈95% of fecal radioactivity, whereas the parent compound accounted for the remainder.

The data on the levels of metabolites in urine and feces (expressed as % of dose) were summarized in the following sponsor's table:

Table 4. Percentages of metabolites of CP-88,059-01 in human subjects after oral administration of [^{3}H]- and [^{14}C]-CP-88,059-01

Metabolite #	% of Dose[a]		
	Urine[b]	Feces[c]	Total
M1+M2[e]	11.25	ND	11.25
M3A[f]	5.38	ND	5.38
M4[f]	6.93	ND	6.93
M4A[f]	0.37	ND	0.37
M5+M6+M7[e]	1.51	ND	1.51
M8+M9+M10[e]	6.16	63.05	69.21
M13	0.00	3.25	3.25

a; average of (^{3}H) and (^{14}C) labels.
b; based on 20.59% (^{3}H) and 20.07% (^{14}C) recovery.
c; based on 68.11 (^{3}H) and 64.52% (^{14}C) recovery.
d; only (^{3}H) label.
e; not separated on
f; only (^{14}C) label.

In serum, 10 peaks were detected. The following peaks were identified: Peak 1 (M1), Peak II (M2), Peak III (M3), Peak IV (M3A), Peak V (M4), Peak VI (M4A), Peak VII (M5), Peak VII (M6), Peak VII (M7), Peak VIII (M8), Peak IX (M9), Peak IX (M10), Peak X (M13). The parent compound accounted for 24-28% of serum radioactivity. The major serum metabolite appeared to be M2, a cleaved molecule, which accounted for ≈22% of serum radioactivity. M9 + M10 accounted for 14-18% of serum radioactivity. Identified metabolites and parent compound accounted for 58-70% of drug-related material in serum.

The proposed pathways for the metabolism of CP-88,059 in humans is summarized in the sponsor's Figures 33-35 (provided below). The major pathways were as follows:

(1) "...N-dealkylation of ethyl side chain attached to piperazinyl nitrogen (M4 and M5)..."

(2) "...oxidation on the benzisothiazole moiety (other than sulfur, M6 and M9)..."

(3) "...hydroxylation on the 3-position and subsequent oxidation of the benzisothiazole moiety (M4A and M7)..."

(2) and (3) are considered to be unusual pathways for this class of drugs.

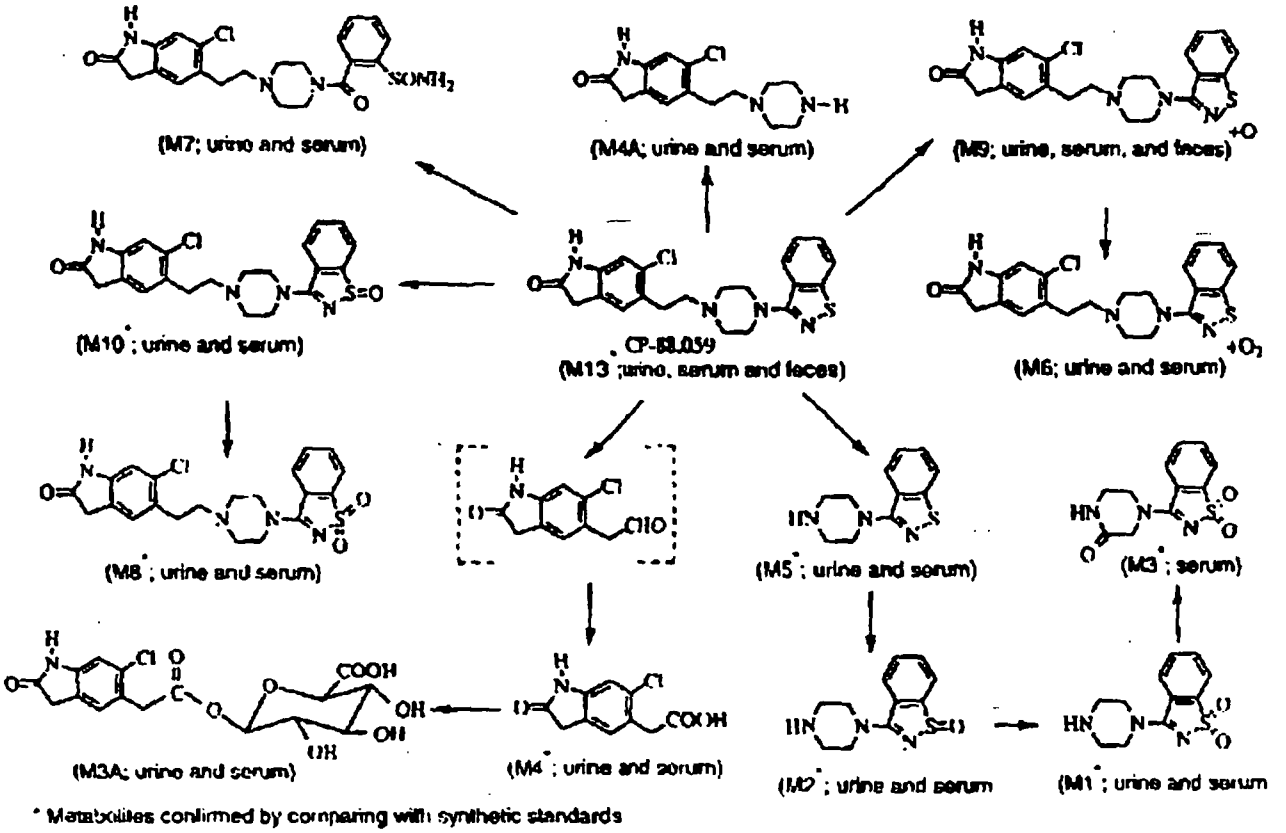


Figure 33. Proposed routes for the biotransformation of CP-88,059 in man

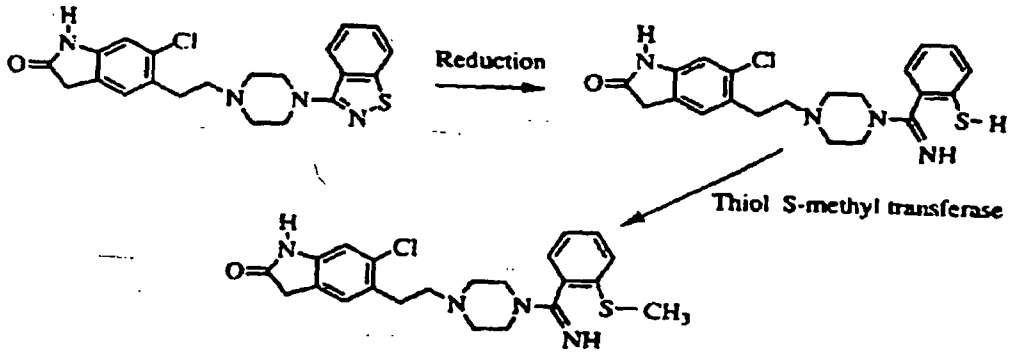


Figure 34. Proposed pathway for the formation of metabolite M9

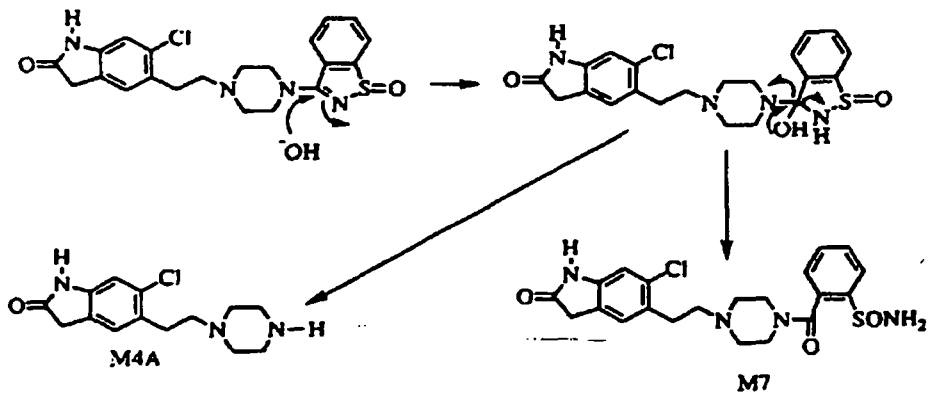


Figure 35. Mechanism for the formation of metabolites M4A and M7

4. **Determination of enzymes involved in the metabolism of ziprasidone** (Study no. DM 95-128-29).

Methods: [^{14}C]-CP-88,059 was used in this series of experiments. [^{14}C]-CP 118,954 was used as the internal standard. Experiments were conducted in human hepatic microsomes from human organ donor tissue. Each microsomal preparation was characterized for the following (as listed by the sponsor): total protein content, total P450 content, mephenytoin 4'-hydroxylation (MP), tolbutamide hydroxylation (TB), bufuralol 1'-hydroxylation *BUF), testosterone 6 ß-hydroxylation (TES), phenacetin)-deethylation (PHEN) activities. Enzyme kinetics was tested in microsomes prepared from a liver sample from one human subject.

The following experiments were performed:

(1) substrate saturation.
(2) time course for formation of ziprasidone-sulfone and ziprasidone-sulfoxide at 10 and 100 µM. Reaction sampling times were 0, 5, 15, 30, 45, 60 min of incubation.
(3) formation of the sulfone and sulfoxide metabolites was tested in microsomal preparations from 8 individuals.
(4) inhibition studies using ketoconazole (10 µM).

Reactions were stopped by the addition of 2 mL of methanol. Microsomal metabolites were quantitated using The limits of quantitation were to µg/mL.

Results: 3-4 major metabolites were detected following incubation of [^{14}C] CP 88,059 with all microsomal preparations. Formation of the sulfone and sulfoxide metabolites was linear up to 5 min, with little additional formation after 30 min. The apparent V_{max} for formation of sulfone + sulfoxide was 1.14 nmol/mg protein/min; the apparent K_M was 235 µM. The intrinsic microsomal clearance of ziprasidone-sulfoxide (i.e., sulfone + sulfoxide) was 0.0048 mL/mg protein/min.

In microsomal preparations from 7 humans, the rate of formation of the sulfoxide metabolites was correlated with activity of TES (CYP3A4, r = 0.83) and MP (CYP 2C19, r = 0.7). The involvement of CYP3A4 was confirmed using ketoconazole. Ketoconazole added to the

incubation medium inhibited formation of ziprasidone sulfoxides by 77%. However, the sponsor noted that the formation of the ziprasidone-desethylene metabolite was unaffected by ketoconazole, suggesting that the formation of this metabolites does not involve CYP3A4.

The sponsor concluded that liver cytochrome P-450 enzymes (particularly CYP3A4) are involved in metabolism of ziprasidone. The two major pathways identified from these experiments are as follows: (1) oxidation at the sulfur of the benzisothiazole and (2) oxidation on the piperazine ring. The sponsor noted that the sulfone and sulfoxide metabolites are major plasma metabolites in humans.

The pathways for formation of the products of *in vitro* metabolism of ziprasidone by human hepatic microsomes were summarized in the following sponsor's Figure 4:

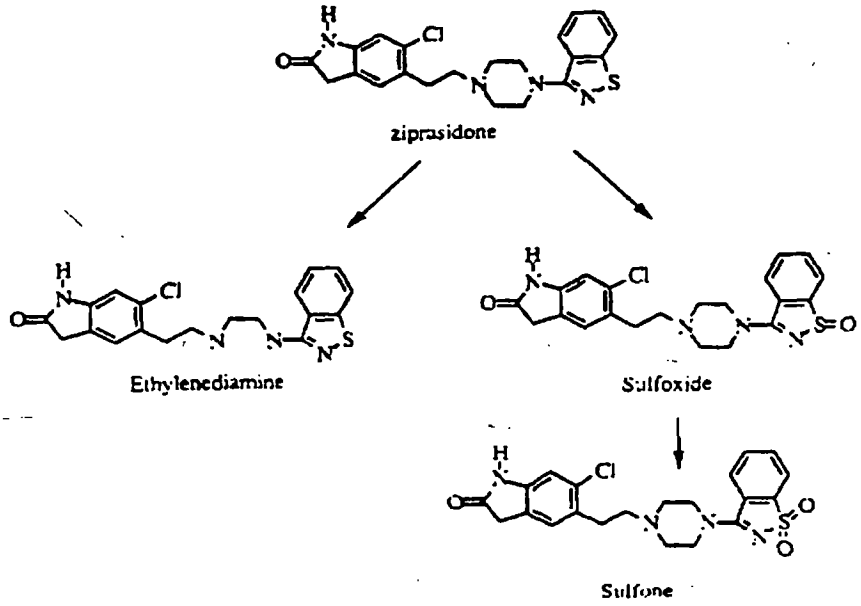


Figure 4. Major *in vitro* Metabolites of ziprasidone with hepatic microsomes

5. **Interaction of ziprasidone and risperidone with human cytochromes P450** (Study no. DM-94-128-21).

Methods: inhibition by CP 88,059 and risperidone (0.1-100 μM) of the activity of five major drug metabolizing enzymes (CYP P4501A2 (CYP1A2, phenacetin O-deethylase), CYP2C9 (tolbutamide hydroxylase), CYP2C19 (S-mephenytoin hydroxylase), CYP2D6 (bufuralol 1'-hydroxylase), and CYP3A4 (testosterone 6ß-hydroxylase) was tested in human liver microsomes from 3 individuals. IC_{50}'s were estimated by curve fitting and reading off the concentration at 50% activity. K_i's were calculated only when the IC_{50} estimate was <100 μM, using the equation: $IC_{50} = (1 + [S]/K_m)K_i$.

Results: neither ziprasidone or risperidone inhibited activity of CYP1A2, CYP2C9, or CYP2C19 (i.e., IC_{50} > 100 μM). Both compounds inhibited CYP2D6 and CYP3A4 with similar potency. K_i's for CYP2D6 were 5.8 ± 4.8 and 4.5 ± 2.9 μM, respectively. K_i's for CYP3A4 were 64 ± 22 and 67 ± 15 μM, respectively.

The sponsor estimated the free plasma concentration in humans at a dose of 80 mg b.i.d. of

ziprasidone to be 0.29-0.73 nM and at 8 mg b.i.d. of risperidone to be 0.24 μM.

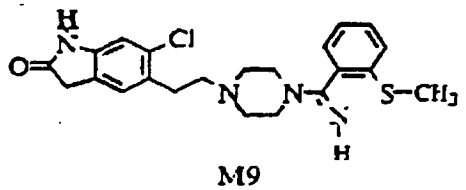
Interspecies comparisons

1. **Further characterization of rat, mouse, dog, and human CP-88,059 metabolites M6 and M9** (Study no. DM-95-128-27).

Methods: M6 and M9 were characterized in urine, bile, and feces samples. Urine samples were collected from male mouse, rat, dog, and human volunteer. Bile samples were collected from rat. Feces were obtained from human subjects enrolled in a balance study. Metabolites were quantitated using

Results: M6 and M9 were found in samples from all species tested, but were most abundant in urine and bile from rat and in human feces. Therefore, characterization of M6 and M9 was conducted using these sources.

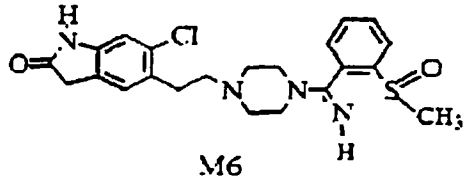
M9 was tentatively identified as S-methyl-CP-360,089 (structure provided by sponsor):



M9

This structure was confirmed by co-elution with authentic standard CP-374,264.

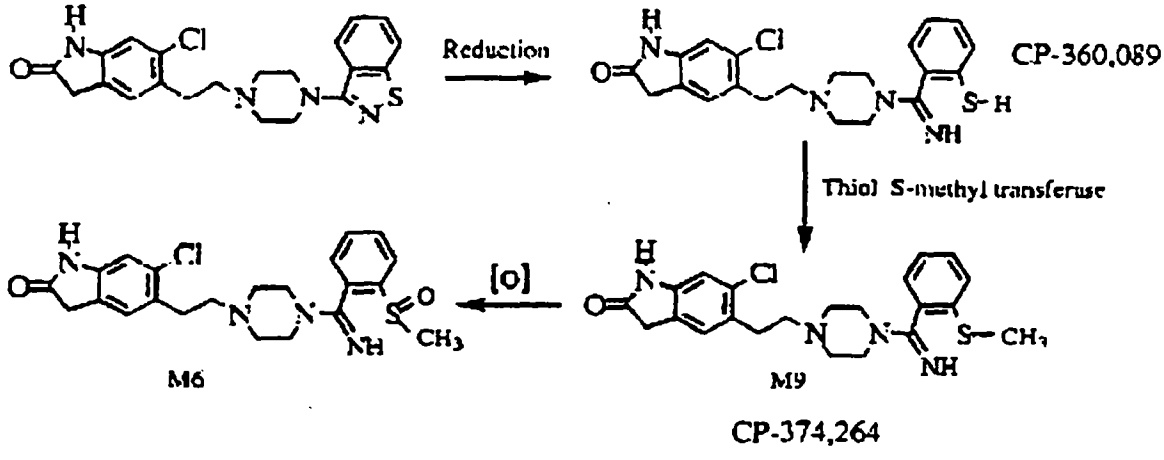
M6 was tentatively identified as CP 360,089-S-methyl-S-oxide (structure provided by sponsor):



M6

M6 did not co-elute with any of the authentic standards.

The proposed pathways for formation of M6 and M9 were summarized by the sponsor in the following figure:



Proposed pathway for the formation of M6 and M9

Protein binding

1. **Binding characteristics of CP-88,059 to melanin** (Protocol No. 94B-88,059-GEN 1,2, 1).

Methods: binding of [^{3}H] CP-88,059 to synthetic dopamine melanin (MW is unknown) and in 10% bovine retina homogenate was tested *in vitro*. Identification of binding sites was examined using Scatchard plots (B/F vs B).

Results: approximate 92-93% of the [^{3}H] CP-88,059 added bound to both synthetic melanin and the bovine retina homogenate. Washing with 50% ethanol resulted in removal of the majority (≈98%) of bound drug, indicating reversibility of binding. The Scatchard plot revealed two classes of binding sites.

Although no data or details were provided, the report stated that changes in ionic strength of the medium had no effect on binding, suggesting hydrophobic interaction.

2. **Protein binding of CP-88,059 to human albumin and α_1-acid glycoprotein and the interaction of CP-88,059 with warfarin and propranolol** (Study no. DM-96-128-36).

Methods: plasma protein binding of CP-88,059 was determined by equilibrium dialysis To test the effects of CP-88,059 on binding of warfarin or propranolol, CP-88,059 (lot no. 20480-71-2MS) was tested at concentrations of 0, 200, and 400 ng/mL. The effects of [^{14}C] warfarin (0, 7.4, 74 µg/mL) and[^{3}H] propranolol (50 ng/mL) on CP-88,059 binding was also tested. [Warfarin binds mainly to albumin and propranolol binds primarily to AAG.] Human plasma was used for all binding assays. Bound The sponsor noted that low concentrations of CP-88,059 were detected in samples of buffer. Therefore, was used to quantitate unbound CP 88,059 in buffer. was used to quantitate unbound CP 88,059 in plasma samples.

Results: plasma protein binding of CP 88,059 was high (>99%) and was not affected by either warfarin or propranolol. Similarly, CP 88,059 had no effect on the plasma protein binding of either warfarin or propranolol, both of which were highly bound (>99 and 91%, respectively).

In a separate assay, binding of CP 88,059 to AAG and albumin were determined to be 98.21 ± 1.41 and 97.96 ± 0.39%, respectively.

3. **Binding of CP-88,059 in monkey and human plasma** (Study no. DM95-128-31).

Methods: plasma protein binding was determined by equilibrium dialysis
Cebus monkey samples were from 6 animals and were pooled for analysis. The cynomolgus monkey sample was purchased. Human serum samples were obtained in-house. Samples were analyzed using either or
The LLOQ were ng/mL and pg/mL, respectively. The sponsor noted loss of data in 4/5 dialysis cells using monkey plasma due to methodological difficulties. No explanation was given for why these cells were not repeated.

Results: plasma protein binding was high in both species, 99.68, 99.83 ± 0.02, and 99.91 ± 0.005% in cebus monkey, cynomolgus monkey, and human plasma, respectively.

The cebus monkeys had been given a 1.5 mg/kg i.m. dose of CP-88,059. These animals exhibited dystonia from 15 min postdosing on. Blood samples were collected at 20 and 45 min postdosing, and the mean "C_{max}" (20 min sample) was 345 ng/mL. In patients receiving 80 mg b.i.d. in clinical trials, no dystonia was reported. It was concluded that the difference in the free fraction between the two species was responsible for this discrepancy. The data were presented in the following sponsor's table:

Table 2
Summary of Mean Protein Binding Data

Species	Dose	Fb (%)	Fu (%)	Total Cmax* (ng/ml)	Free Cmax (ng/ml)
Monkey:					
cebus	0.5 mg/kg IM	99.68	0.32	345	1.104
cynomolgus	0.5 mg/kg IM	99.83	0.17	NA	NA
Human	80 mg BID	99.91	0.09	250	0.225

* - Total Cmax - bound and free concentration of drug
NA - not applicable

First, there is a discrepancy in the dose given to monkeys between the text and the table. According to the text, the dose was 1.5 mg/kg, but in the table the dose was listed as 0.5 mg/kg. Second, it must be remembered that the % plasma protein binding data were based on a sample from one animal in cebus monkey and on samples from two humans. Third, the data in cebus monkey was based on a 20-min sample; after an i.m. injection, the true C_{max} may have occurred prior to this time.

Acute

The acute toxicity of CP-88,059 was tested in albino mice and Sprague-Dawley rats. This study was previously reviewed (P/T review, 4/13/90, J. DeGeorge, Ph.D.). The following summary is based on this review.

CP-88,059 was administered to mice (strain not specified) at doses of 500 and 2000 mg/kg p.o. (3/sex/grp) and 300, 500, and 1000 mg/kg i.p. (3 M only). Rats received doses of 500 and 2000 mg/kg p.o. (3/sex) and 500 and 2000 mg/kg i.p. (3 M only). Drug-related deaths occurred only in male mice at 1000 mg/kg i.p. Sedation was the primary clinical sign with both routes. CNS signs tended to have a more rapid onset and prolonged duration with i.p. dosing. No target organ for toxicity was identified.

LD_{50}'s were calculated to be >2000 mg/kg p.o. in both mice and rats and >2000 mg/kg i.p. in rats, and 500-1000 mg/kg i.p. in mice.

This study was not definitive due to the lack of a complete battery of measurements, of control grps, and the small n/grp.

Subchronic

Subchronic oral toxicity studies were conducted in Long-Evans rats (2-wk, 1-mo) and Beagle dogs (2-wk, 1-mo). The 2-wk study in rat and the 1-mo studies have been previously reviewed (2-wk rat: P/T review, 10/28/92, L.M. Freed, Ph.D.; 1-mo studies: P/T review, 4/13/90, J. DeGeorge, Ph.D.). The following summary of these studies are based on these reviews, with reference to the original reports as needed. The original report was examined for the 2-wk study in dogs. The TK data for these studies are summarized in attached tables.

Rat: the 2-wk study was conducted at doses of 0, 5, 25, and 75 mg/kg (gavage). This study was not definitive due to limited histopathology and the small n (3/sex/grp). The 1-mo study was conducted at doses of 0, 10, 40, and 160 mg/kg (gavage, 10/sex/grp). The report of the 1-mo study was incomplete due to the lack of a complete summary histopathology table.

There were no unscheduled deaths in either study. The primary drug-related clinical sign was sedation (ptosis, reduced motor activity), which was noted at all doses in both studies. Body weight was reduced (compared to controls) at the MD and HD in the 2-wk study (6-8 and 8-15%, respectively) and at all doses in the 1-mo study (≈10-25%). In the latter study, body weight loss was noted in males at the MD and HD during the first 2 wks of dosing. In females, body weight was minimally affected in the 1-mo study at the LD and MD (3-4%), but was 8% lower than CF at the HD. Changes in food consumption in males were fairly consistent with those in body wt in the 1-mo study; in females, food consumption tended to be less affected, if at all. No effects were noted on hematology, urinalysis, ophthalmology (1-mo study), or gross pathology in either study. In the 1-mo study, ALT was slightly elevated in males and females at the HD (37-70%). A complete battery of organ weights were not collected (1-mo study); only kidney, liver, and testes were weighed. No drug-related changes were noted, except those apparently related to changes in body wt. Microscopic analysis of tissues was performed only in C and HD animals, and only a limited summary table was provided. No apparent drug-related findings were noted.

Dog: the 2-wk study was conducted at doses of 0, 2, 5, 10, and 20 mg/kg (gavage). This study was not definitive due to the small n (1/sex/grp) and the lack of terminal studies. The 1-mo study was conducted at doses of 0, 10, 20, and 40 mg/kg (gavage). C and LD grps were given

once/daily dosing, whereas the MD and HD were administered b.i.d. (i.e., 10 and 20 mg/kg b.i.d.) with ≈4 hr between doses. The n in the 1-mo study (3/sex/grp) was minimal.

There were no unscheduled deaths in either study. Sedation was the primary clinical sign in both studies, with severity being dose-related. In the 2-wk study, the sponsor noted that other signs (e.g., splayed hindlimbs, intermittent tremors, ptosis, and shallow breathing) were independent of sedation. No effects on body weight, food consumption, ECG, hematology, urinalysis, or histopathology were noted. In the 1-mo study, however, ECG data were not provided and it was not clear that quantitative data were collected. Increases in ALT were noted at the MD and HD in the 1-mo study (2.5-3.8 fold); one HDF had increases in both ALT and AST (2-3 fold). A complete battery of organ weights were not collected (1-mo study); only kidney, liver, and testes were weighed. No drug-related effects were noted in these organs.

Chronic

Chronic toxicity studies were conducted in Sprague-Dawley rat (6-mo) and Beagle dog (6-mo, 1-yr). All three studies were conducted under GLP and have been previously reviewed (6-mo rat: P/T review, 10/28/92, L.M. Freed, Ph.D.; 6-mo dog: P/T review, 2/16/93, L.M. Freed, Ph.D.; 1-yr dog: P/T review, 10/6/94, L.M. Freed, Ph.D.). The following summary is based on these reviews, with reference to the original reports as needed. The TK data for these studies are summarized in attached tables.

Rat: the 6-mo study was conducted in 15/sex/grp at oral (gavage) doses of 0, 10, 40, and 200 mg/kg. Measurements consisted of the following: clinical signs, ophthalmology, clinical pathology [hematology (wbc ct, differential, rbc, hgb, hct, MCH, MCHC, MCV, platelet ct, fibrinogen), clinical chemistry (Na, K, Ca, Cl, BUN, creatinine, ALT, AST, SDH, total protein, albumin, 5'-nucleotidase, cholesterol, TG, total bilirubin), urinalysis (specific gravity, volume, pH, protein, blood, glucose, urobilinogen, bilirubin, ketones], and terminal studies [organ/tissue wts (kidneys, liver, testes), gross pathology, histopathology (list of tissues in appendix)]. Microscopic analysis of tissues was performed in C and HD grps; the LD and MD grps were examined only if a potentially drug-related finding was observed at the HD.

There were 11 unscheduled deaths. Of these, 8 were attributed to trauma during dosing or upon recapture (i.e., handling error). The cause of death in 1 CM, 1 LDF, and 1 HDM could not be determined. The primary drug-related clinical sign was sedation (to the point of unconsciousness) which was noted at all doses. One sign noted only at the HD was described as a peculiar tail reflex consisting of arching of the tail over the back. Difficulty in handling and food wastage also were limited to the HD grps. Body weight was affected in both males and females. In males, body weight loss was noted during the first wk of dosing (-5.4 gm vs. +47.0 gm in CM). Final mean body weight was reduced (compared to CM) at all doses in males (≈17, 31, and 35% in LDM, MDM, and HDM, respectively). In females, final mean body weight (compared to CF) was slightly elevated at the LD (3.5%), but reduced at the MD and HD (6 and 9%, respectively). Food consumption could not be accurately assessed at the HD due to instances of food wastage. In the lower dose grps, food consumption was reduced in males, but not females. There were no drug-related findings on ophthalmology (3, 6-mo); however, repeated orbital sinus bleeding may have compromised the evaluation. On hematology parameters, decreases were noted in wbc ct at the HD (37-20%), with the differential analysis indicating an increase in neutrophils (≈70%) and a decrease in lymphocytes (10-12%). The only notable findings in clinical chemistry parameters were increases in ALT (55-60% at Day 194), AST (81 and 45% at Day 194), and 5'-nucleotidase (.36% at Day 194 in males only) at the HD. No drug-related findings were noted on urinalysis parameters. A complete battery of organ/tissue weights were not performed; only kidneys, liver, and testes were weighed. Changes in kidney and liver wt reflected decreases in body weight. Absolute testis wt, however, was similar among grps, reflecting either sparing of testis from body weight-induced changes or an increase in wt masked by reduced body weight gain. According to the sponsor, there were no gross findings at necropsy (no summary or line

Long-Evans rat, TK data summary

STUDY	N	SAMPLING TIME	SEX	DOSE (mg/kg)	C_{max} (µg/mL)
2-wk	3/sex/grp	Day 1	M	5	0.76*
				25	2.54
				75	3.36
			F	5	1.06
				25	2.25
				75	5.22
		Day 14	M	5	0.97
				25	3.12
				75	4.68
			F	5	1.27
				25	3.26
				75	6.40
1-mo	5/sex/grp	Day 1	M	10	0.97**
				40	3.13
				160	4.61
			F	10	0.75
				40	2.65
				160	6.08
		Day 10	M	10	0.87
				40	3.03
				160	9.21
			F	10	1.08
				40	3.34
				160	11.02
		Day 26	M	10	0.80
				40	3.52
				160	9.24
			F	10	1.35
				40	3.88
				160	9.78

*1-hr postdosing sample, **based on 1 and 2-hr postdosing samples

Long-Evans rat, TK data summary, con't

STUDY	N	SAMPLING TIME	SEX	DOSE (mg/kg)	C_{max} (μg/mL)
6-mo	5/sex/grp	Day 10	M	10	0.74
				40	3.55
				200	8.86
			F	10	0.85
				40	2.70
				200	11.66
		DAY 89	M	10	1.14
				40	6.22
				200	9.40
			F	10	1.45
				40	5.13
				200	9.46
		DAY 173	M	10	1.0
				40	3.96
				200	6.86
			F	10	1.29
				40	3.10
				200	10.32

*1-hr postdosing sample, **based on 1 and 2-hr postdosing samples-

Beagle dog, TK data summary

STUDY	N	SAMPLING TIME	SEX	DOSE (mg/kg)	C_{max} (μg/mL)	AUC (μg•hr/mL)
2-wk	(1/sex/grp)	Day 1*	M + F	2	0.34	1.4#
				5	0.47	2.1
				10	0.73	3.3
				20	0.57	2.8
		Day 9		2	0.35	1.4
				5	0.72	1.9
				10	1.37	5.0
				20	0.57	2.6
		Day 10		20	0.57	2.6
1-mo	3/sex/grp	Day 2	M + F	10	0.20	
				20	0.74	
				40	2.56	
		Day 8		10	0.58	
				20	0.92	
				40	0.73	
		Day 23		10	0.83	
				20	0.54	
				40	0.58	

*Day 1 values were not collected on Day 1 due to methodological errors, but were collected "...following a one week wash-out period"; the actual day of collection was not specified.
#blood samples were collected for up to 6 hr postdosing.

TK data from 6-mo oral toxicity study in Beagle dog:

DOSE (mg/kg)	DAY	HR*	PLASMA CONC. (ng/mL)	$AUC_{(0\text{-}12\ hr)}$ (ng•hr/mL)
5	10	2	155 ± 96	685 ± 421
		6	51 ± 36	
	71	2	171 ± 94	
		6	52 ± 39	
	127	2	221 ± 110	
		6	75 ± 32	
	170	2	159 ± 59	
		6	60 ± 22	
10 (5 b.i.d.)	10	2	176 ± 92	1993 ± 789
		6	341 ± 165	
	71	2	296 ± 177	
		6	324 ± 152	
	127	2	322 ± 161	
		6	395 ± 192	
	170	2	231 ± 105	
		6	335 ± 141	
40 (20 b.i.d.)	10	2	720 ± 495	5921 ± 2963
		6	899 ± 440	
	71	2	589 ± 393	
		6	766 ± 384	
	127	2	646 ± 224	
		6	724 ± 283	
	170	2	926 ± 439	
		6	1040 ± 584	

*time from 1st daily dose; when dosing was b.i.d., doses were given ≈4 hr apart

listings were provided for survivors). The primary histopathology findings were as follows: (1) lung changes consisting of pleuritis (a few HD animals) and multifocal granulomatous pneumonia (all grps, but an increase in incidence and severity in 1 MDF and HD animals). This finding was characterized as aggregates of foamy macrophages and was attributed to aspiration of drug during dosing, (2) adrenal gland changes consisting of multifocal cystic degeneration and multifocal telangiectasia (i.e., dilation of small or terminal vessels) in MD and HD females (telangiectasia was also detected in 1 LDF), focal fibrosis in 1 HDF, and diffuse hypertrophy (characterized by increased cytoplasmic mass in cells of the zona fasciculata in MDF, and HD animals. (3) multifocal, suppurative prostatitis in MD and HD males. In terms of the adrenal changes, the sponsor suggested that adrenal cortical telangiectasia and cystic degeneration may "...represent different stages of the same degenerative process...Cystic degeneration is....believed to occur secondary to lesions in the cortical capillaries." Both findings were detected together in 1 MDF and 3 HDF; however, in 1 MDM, 3 MDF, 1 HDM, and 3 HDF only one or the other was found. Selected microscopic findings are summarized in the following table:

TISSUE	FINDING	CM	LDM	MDM	HDM	CF	LDF	MDF	HDF
lung	granulomatous pneumonia, multifocal	1/15	0/15	2/15	8/15	2/15	2/15	3/15	4/15
adrenal	hypertrophy, diffuse (z. fasiculata)	0/15	0/15	0/15	7/15	0/15	0/15	5/15	8/15
	telangiectasia, multifocal	0/15	0/15	0/15	0/15	0/15	1/15	4/15	6/15
	cystic degeneration, multifocal	1/15	0/15	1/15	1/15	2/15	0/15	1/15	4/15
prostate	prostatitis, multifocal, suppurative	7/15	5/15	13/15	13/15				

Dog: 6-mo and 1-yr oral toxicity studies were conducted in Beagle dog. Both of these studies have been previously reviewed (6-mo: P/T review, 2/16/93, L.M. Freed, Ph.D.; 1-yr: P/T review, 10/6/94, L.M. Freed, Ph.D.). The following summary is based on these reviews, with reference to the original report as necessary.

The 6-mo study was conducted at doses of 0, 5, 10, and 40 mg/kg; dosing was by gavage. The MD and HD were given as 5 and 20 mg b.i.d., respectively. There were 4 dogs/sex/grp. Measurements consisted of the following: clinical signs, ECG/vital signs, ophthalmology, hematology (wbc, rbc, hgb, hct, wbc differential, platelet ct, APTT, PT, MCV, MCH, MCHC), clinical chemistry (Na, K, Ca, Cl, glucose, BUN, creatinine, ALT, AST, alk phos, SDH, GGTP, total protein, total bilirubin, albumin, cholesterol, TG), urinalysis (volume, color, specific gravity, pH, protein, blood, glucose, urobilinogen, bilirubin, ketones), TK, terminal studies [gross pathology, organ/tissue wts (kidneys, liver, testis), and histopathology].

There was one unscheduled death during the study. One HDM was sacrificed during Wk 12 after fracturing several teeth and the jaw during cage-biting; this behavior was considered drug-related by the sponsor. Drug-related clinical signs were noted in all drug-treated dogs, and consisted of reduced motor activity, recumbency, leaning/pressing against the cage, pawing, limb extension/unusual posture, tremors, hypersalivation, ptosis, and panting. Signs noted at all doses, but not necessarily in all drug-treated animals included vocalization,increased activity, pacing/circling, aggressive behavior toward humans, cage biting, muscle fasciculation, emesis, and prolapse of nictitating membrane. Body weight gain was reduced (compared to C) in males at all doses, in a dose-related manner; increases in body weight of CM, LDM, MDM, and HDM by the end of the study were 26, 16, 9, and -2%, respectively). In females, there were no dose-related changes in body wt; body weight gain was reduced only in MDF. Food consumption was not affected by drug-treatment, i.e., changes in body weight were not reflected in changes in food consumption. There were no drug-related changes in ECG parameters, systolic blood pressure, or rectal temperature. Miosis, blepharospasm, and prolapse of the nictitating membrane were noted upon

ophthalmology examination. Miosis was noted at the MD and HD, and the latter findings were time- and dose-dependent; blepharospasm was noted at all doses, whereas, prolapse of the nictitating membrane was observed in MD and HD animals. There were no clear drug-related findings on hematology or urinalysis parameters. On clinical chemistry parameters, drug-related findings consisted primarily of increases in alk phos and ALT at the HD. The effect on these parameters increased with duration of dosing, with high values being detected in alk phos in 4/8 HD (243-404 U/L) and in ALT in 8/8 HD (58-183 U/L). There were no drug-related effects on kidney, liver, or testis wt not accounted for by changes in body weight. The only gross lesions noted at necropsy were self-inflicted injury in 1 HDM (i.e., fractured teeth, maxilla) resulting from aggressive, cage-biting behavior. Drug-related histopathology was detected in liver and kidney. Intrahepatic cholestasis, characterized by the sponsor as "...bile plugs within canaliculi and phagocytized bile within Kupffer cells...", was noted in all HD animals. No evidence of hepatic necrosis or inflammation was noted. Accumulation of lipofuscin was detected in the renal proximal tubules, with the incidence and severity being dose-related (1/4 CF, 3/4 MDM, 2/4 MDF, and all HD animals).

The 1-yr study was conducted at doses of 0, 5, 10, and 20 mg/kg. Dosing was by gavage and the MD and HD were given as 5 and 10 mg/kg b.i.d. There were 4 dogs/sex/grp. Measurements consisted of the following: clinical signs, physical examination (heart rate, respiratory rate, rectal temperature), body weight/food consumption, ophthalmology, ECG/blood pressure, hematology (rbc, hgb, hct, wbc ct, wbc differential, platelet ct, MCH, MCH, and MCHC), clinical chemistry (Na, K, Ca, Cl, BUN, creatinine, alk phos, total bilirubin, bile acids, ALT, AST, SDH, total protein, albumin, GGTP, glucose, cholesterol, TG, globulin and A/G ratio were calculated), urinalysis (volume, specific gravity, pH, protein, blood, glucose, urobilinogen, bilirubin, ketones, color), TK, and terminal studies [gross pathology, organ/tissue wts (kidney, liver, and testis), histopathology].

There were no unscheduled deaths during the study. Drug-related clinical signs were evident at all doses and were listed by the sponsor as "... ptosis, tremors, recumbency, head pressing, pawing, unusual postures, increased and/or decreased activity, aggressive behavior, cage biting/licking, muscle fasciculations, ataxia, rapid respiration, and vocalization". Aggressive behavior was so severe in 1 HDM (# 27) that the second daily dose was discontinued in this animal from Day 130 on. No drug-related effects were noted on mean body weight, food consumption, physical examination/vital signs, ECG/blood pressure, ophthalmology, hematology, clinical chemistry, or urinalysis parameters. Increases in ALT (2-3 fold), however, were noted in individual animals (1 CM, 2 LDM, 3 HDM, 2 HDF), but were not dose-related on the final measurement day (i.e., Day 357-360). There were also no drug-related findings on any of the terminal studies, including histopathology.

Special studies

1. **Exploratory prolactin study in rats** (Study no. 94-720-29, Pfizer Central Research, report date: 10/95, non-GLP, Vol. 1.27).

Methods: ziprasidone (lot no.) was administered to female Long-Evans rats (20/grp) at doses of 0, 2, 6, and 12 mg/kg p.o. (dietary) for ≈5 wks. According to the sponsor, experimental conditions were "...identical to those employed in a 2-year bioassay....(Study #92-720-21)". The following observations were made: (1) serum prolactin levels were quantitated after 2 and 4 wks of dosing, (2) stage of estrus cycle was determined daily by vaginal lavage, (3) plasma ziprasidone levels were quantitated on Day 38 (≈1 p.m.; 10/grp) or Day 39 (1:00 a.m.; 10/grp); these sampling times were intended to capture trough and peak levels). Serum prolactin was quantitated by using antibodies to rat prolactin. Plasma ziprasidone was quantitated using .

Results: there were no differences in serum prolactin levels at any dose. The data (taken directly from sponsor's Table 1; mean ± SD, expressed in ng/mL) are summarized in the

following table:

DAY	CF	LDF	MDF	HDF
-7	37.88 ± 70.028	18.20 ± 16.332	23.50 ± 18.924	24.89 ± 26.644
15	33.41±27.317	31.57 ± 24.251	37.56 ± 28.612	36.83 ± 35.352
29	73.85 ± 40.632	51.44 ± 66.096	69.85 ± 83.525	62.94 ± 42.440

Plasma levels of ziprasidone were as follows [means ± S.D. (ranges), expressed as ng/mL; data taken directly from sponsor's tables]:

SAMPLING TIME	LDF	MDF	HDF
1:00 A.M.	56.9 ± 13.5 (39.8-80.5)	140.4 ± 28.3 (87.5-186.0)	407.5 ± 77.3 (314.3 ± 556.2)
1:00 P.M.	14.0 ± 8.7 (<5-24.7)	61.9 ± 19.5 (26.4-83.9)	101.3 ± 65.6 (25.6-246.7)

Estrus cycle data are summarized in the following table (taken from sponsor's text table):

OBSERVATION	CF	LDF	MDF	HDF
estrus cycle length (days)	4.3 ± 0.6	4.5 ± 0.5	5.4 ± 1.4	6.7± 1.9
cycles/animal*	6.3	5.9	3.7	1.6

*during the dosing period (Days 1-32)

According to the sponsor, only 1 HDF was cycling normally. Nine HDF cycled only 1-3 times during the dosing period, and 9 HDF were in a continuous state of estrus or diestrus.

2. **1 month in-feed study in CD-1 mice** (Study No. 95-720-36, Pfizer Central Research, study dates: 10/23/95-11/21/95, report date: 3/96, GLP, Vol 1.22)

Methods: ziprasidone (lot no. 31,081-79-1F) was administered to CD-1 mice (15/sex/grp) at doses of 0, 50, 100, and 200 mg/kg/day in the diet (
for 29 days; animals were housed singly. [According to the sponsor, stability and homogeneity of drug in diet were established during Wks 1-4. All admixtures were reported to be within ±15% of intended. Data documentation was not provided.] Observations consisted of the following: clinical signs, body weight, food consumption, serum prolactin (Day 29, using trunk blood at necropsy; method), gross pathology (only ovaries, uterus, and vagina were retained and fixed for "possible future microscopic evaluation").

Results: there were three unscheduled deaths during the study; all were HDF (#108, 1100 on Day 7, #114 on Day 9). Deaths were considered drug-related by the sponsor, and were attributed to "...decreased activity associated with the high dose of 200 mg/kg/day and the resulting dehydration noted in these animals". Drug-related clinical signs consisted of the following: decreased activity (all doses, noted on Days 5-10 only), hunched posture (HDF, Wk 1), and dehydration ("...secondary to decreased activity...") was noted in 6/15 HDF; three of these HDF died.

Body wt was reduced in MDM and HDM (compared to CM) throughout the dosing period (6-7%

and 8-11%, respectively), and in HDF (compared to CF) only during the first wk of dosing (9%). Effects on food consumption were consistent with those on body wt. Mean drug intakes were 88-111, 86-106, and 91-108% of intended. The serum prolactin data (ng/mL; mean ± SD) are summarized in the following table:

SEX	C	LD	MD	HD
M	16.04 ± 6.747 (6.8-29.8)#	11.77 ± 3.640* (7.5-22.1)	19.25 ± 15.727 (5.4-60.3)	18.24 ± 14.509 (6.8-66.5)
F	45.38 ± 116.635 (4.8-465.8)	83.93 ± 96.338 (2.8-339.4)	187.27 ± 206.267* (11.6-772.3)	197.08 ± 134.634** (10.0-405)

#no. in parentheses denote range; *p < 0.05, **p < 0.01

3. **Antigenicity study report** (Protocol No. 93-03-81, Pfizer Pharmaceuticals, Japan, study dates: 2/19/93-4/14/93, report date: 12/93, Japanese GLP, Vol 1.29).

Methods: the antigenic potential of ziprasidone (lot no. 21576-181-1F, 0.5% methylcellulose or water for injection) was tested in the active systemic anaphylaxis (ASA) and the homologous passive cutaneous anaphylaxis (PCA) assays in male guinea pigs.

For the sensitization phase, ziprasidone was administered orally at doses of 3 (= intended clinical daily dose) and 15 mg/kg and subcutaneously at doses of 2 and 10 mg/guinea pig (50 µg/mL = HC). For the challenge dose, ziprasidone was administered at a dose of 100 µg/guinea pig. BSA (1 ng/guinea pig) was used as a positive control.

For the ASA assay, ziprasidone was administered orally (1 dose/day, 5 days/wk) for 3 wks or s.c. (4 doses at weekly intervals) during the sensitization phase. Challenge doses (ziprasidone, 50 or 100 µg/guinea pig; saline, or BSA) were administered 19 days after the last oral dose or 16 days after the last s.c. dose. Animals (5/grp) were observed 24 hr after the challenge dose for anaphylactic symptoms (e.g., weakness, labored breathing, sneezing, rales, convulsion, prostration, death) according to the following scale: 0 = asymptomatic, I = 1-3 symptoms (mild), II = 4-7 symptoms (moderate), and .III = >8 symptoms or death (serious).

For the PCA assay, blood samples were drawn from animals sensitized during the ASA assay, 2 days prior to the challenge dose. Serial dilutions of these blood samples were injected intradermally into naive animals (2/serum sample). At 4 hr postinjection, animals were challenged with either ziprasidone, saline, or BSA i.v. together with Evans blue dye. Thirty min later, animals were sacrificed and the diameter of the blue area around the injection site was measured. Blue areas of >5 mm in diameter were considered positive responses.

Results: no guinea pig sensitized with ziprasidone, either p.o. or s.c., exhibited any signs of anaphylaxis upon challenge with ziprasidone. In contrast, those sensitized and challenged with BSA exhibited Category I and II (at the LC) and Category III (at the HC) symptoms.

The PCA test was also negative in ziprasidone-treated animals. As in the ASA assay, animals sensitized with serum from BSA-treated animals and challenged with BSA exhibited positive PCA titers (data expressed as "the ratio of the highest dilution of serum at which Evans Blue spot larger than 5 mm in diameter was observed"). The sponsor did not provide raw data, only ratios, for individual animals.

4. **A single dose dermal toxicity study in rabbits and a single dose ocular irritation study in rabbits** (Study No. 92-720-23, Pfizer Central Research, study dates: 9/22/92-9/25/92, report date: 6/12/95, non-GLP?, purity and stability of drug not documented, Vol 1.29).

Methods: to assess dermal toxicity, ziprasidone was applied to intact skin at a single dose of 2000 mg for 24 hrs. Animals (n = 5) were examined 2 days after drug application.

To assess the potential for ziprasidone to produce eye irritation, ziprasidone was applied as a powder (44.9 mg) to the conjunctival sac of the left eye; the right eye served as a control. Animals (n = 3/grp) were evaluated ("...with minimal manipulation and without the use of fluorescein") for 4 days postdosing. Apparently, at 24 hr postdosing, eyes were examined following application of 2% fluorescein.

Both studies were conducted in New Zealand White rabbits.

Results: in the dermal study, no drug-related mortality or clinical signs or changes in body wt or food consumption were observed, nor were any signs if dermal irritation detected. Although the sponsor concluded that ziprasidone is not a corrosive material, the data are inconclusive since no positive control was included in the study. Therefore, the validity of the methodology was not verified.

In the eye study, no signs of ocular irritation were detected in cornea, iris, or conjunctivae according to the summary table. However, according to the text, the following wee noted: (1) redness of the conjunctiva in the treated eye of each rabbit, (2) iritis and "slight circumcorneal reddening" in the eye of one rabbit; in this rabbit. By 48 hrs, all treated eyes appeared normal. Again, as in the dermal study, no positive controls were included in the study; also, no inert substance was tested to control for mechanical irritation effects.

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Two-year carcinogenicity studies were conducted in CD-1 mouse and Long-Evans rat. Dose-range finding (dietary) studies in CD-1 mouse (1- and 3-mo) and Long Evans or Fischer 344 rats (3-wk, 3-mo) and proposed protocols for the definitive studies were submitted for review of dose-selection. These have been reviewed (P/T review, 10/28/92, L.M. Freed, Ph.D.). The carcinogenicity studies have not been previously reviewed.

Mouse: dose selection was based on 1- and 3-mo dose-range finding studies. Both studies used the dietary route. Doses of 0, 50, 100, and 200 mg/kg were proposed and were used in the definitive 2-yr study. The proposed HD appeared to be adequate for male mice based solely on reductions in body weight gain in the preliminary studies. The 1-mo study was a rising-dose study of limited usefulness. In the 3-mo study, body weight was reduced by 3.5, 4.2, and 5.8% in LDM, MDM, and HDM, respectively, on Day 99 of dosing. Body weight was not notably affected in females, with weights being slightly higher at the LD and MD (1.3-1.7%), and only slightly reduced (0.3%) at the HD, as compared to CF. Clinical signs in males and females were minimal. Decreased activity and slightly slow response to stimuli were noted in only a few animals/grp (no discussion of severity or incidence was provided). No hematology, clinical chemistry, TK, or pathology data were provided. It was this reviewer's opinion that the proposed HD (i.e., 200 mg/kg) was adequate for male mice, but that there were insufficient data to determine an MTD in females.

24-month oral (in diet) toxicity and carcinogenicity study in CD1 mice (Study No. 92114, Pfizer Central Research, study dates: 12/92-12/94, report date: 11/2/95, GLP, Vol 1.18-1.21).

Animals: Swiss mice,
initial age: 6 wks
initial body weight: 29.0 ± 1.4 (males) and 23.0 ± 1.2 gm (females)
n = 50/sex/grp except 100/sex/grp for C grps

Drug: CP-88,059-1 (batch nos. 21,576-181-1F, 23,638-95-1F, 31,081-79-1F)
purity: 86.6-88.6% (no Certificate of Analysis was provided)
storage: rm T protected from light
route: oral in diet
drug formulation: drug-diet admixture. The homogeneity of the mixture was tested on the 1st, 3rd, and 5th batches and on batches prepared every 4 wks up to Wk 100. The frequency of preparation was not clearly specified. Homogeneity of mixing was determined on 3 samples/batch; The CV was 0-4%, with actual concentrations being within 10% of intended. Drug concentrations were adjusted weekly. Actual concentrations were within 13% of intended.
doses: final doses were 0, 0, 50, 100, and 200 mg/kg. The initial dose for each of the three treatment grps was 50 mg/kg. This dose was administered for the first 14 days. On Day 15, the MD and HD were raised to 100 mg/kg, and on Day 29, the HD was raised to 200 mg/kg.
duration: dosing was continuous for 720-724 days.

Observations

Clinical signs: animals were observed daily.

Body weight: body weights were recorded prior to dosing, and once weekly during the dosing period.

Food consumption: food consumption was recorded weekly during the first 6 mo of dosing and monthly thereafter.

Water consumption: water consumption was recorded in 20/sex/grp "...over periods of 24 hours from the second month, then once every two months."

Ophthalmology: ophthalmology examinations were performed on 1 C grp/sex and HD animals prior to the start of dosing, every 6-mo during dosing, and at the end of the study in 25/sex/grp examined. Examinations were conducted following induction of mydriasis (tropicamide) and the a Zeiss slit lamp was used to examine the anterior segment of the eye.

Terminal studies

Gross pathology: a complete necropsy was conducted on all animals, including those sacrificed moribund and those found dead (as much as possible).

Organ/tissue wts: wts of the following organs/tissues were recorded: brain, heart, kidneys, liver, testis.

Histopathology: microscopic examination of tissues (list appended) was conducted in all animals. In addition, pituitary sections from selected animals were examined using immunocytochemistry (immunoperoxidase method using rabbit anti-mouse prolactin antibody) in order to better characterize pituitary tumors in these animals (F811, F814, F816, F820, F822, F823, F826, F837, F839, and F848).

Additional examinations were also conducted in one animal, M246, in order to further study apparent hematological changes (i.e., macroscopic): bone marrow smear (femoral) and imprints of cervical lymph node, spleen, thymus. These samples were stained with May-Grunwald Giemsa and myeloperoxidase stains and examined with a light microscope.

Microscopic examination was conducted by two independent pathologists.

Statistics

Survival analysis: each sex was analyzed separately. Survival among grps was compared using the log-rank test. Control grps were analyzed statistically, and were combined for the purpose of analysis if there was no significant difference between grps. If the Cs were significantly different, analyses were performed with each treatment grp being compared to each control separately and combined. Significance was tested at $p<0.05$.

Analysis of pathology findings: observations noted in <5 animals were not statistically analyzed. The following non-neoplastic findings in females were statistically analyzed using the Cochran-Armitage trend test: ovarian atrophy, ovarian cysts, uterine hyperplasia (cystic, endometrial), uterine "inactivity", pituitary hyperplasia (focal, pars distalis), skin and adnexia (galactocele, hyperplasia/lobular, mammary gland).

Neoplastic findings (in ≥5 animals) were analyzed "...using the Peto's death rate method for fatal tumors and prevalence analysis for incidental tumors".

Results

Mortality: there were no significant differences in mortality among grps in either males or females. The survival data are summarized in the following sponsor's table:

Incidence of moribundity and mortality*

		Control 1		Control 2		50 mg/kg		100 mg/kg		200 mg/kg	
		M	F	M	F	M	F	M	F	M	F
Number of animals		50	50	50	50	50	50	50	50	50	50
Sacrificed as moribund		12	10	9	8	9	15	8	10	4	14
Found dead		14	19	12	21	18	14	17	25	16	16
Total unscheduled deaths	No.	26	29	21	29	27	29	25	35	20	30
	%	52	58	42	58	54	58	50	70	40	60
Survivors	No.	24	21	29	21	23	21	25	15	30	20
	%	48	42	58	42	46	42	50	30	60	40

* including the 2 mice found dead during the period of scheduled sacrifice.

Clinical signs: the data were not summarized. According to the sponsor, there were no drug-related findings.

Body weight: body weight was reduced compared to C grps in both males and females. At 200 mg/kg, body weight was 8 and 10% lower in HDM and HDF, respectively, than in combined C grps. Body weight gain was reduced by 8, 19, and 34% in LDM, MDM, and HDM, respectively, and by 10, 33, and 31% in LDF, MDF, and HDF, respectively, at the end of the dosing period. The data are illustrated in the following sponsor's Figures 3 and 4:

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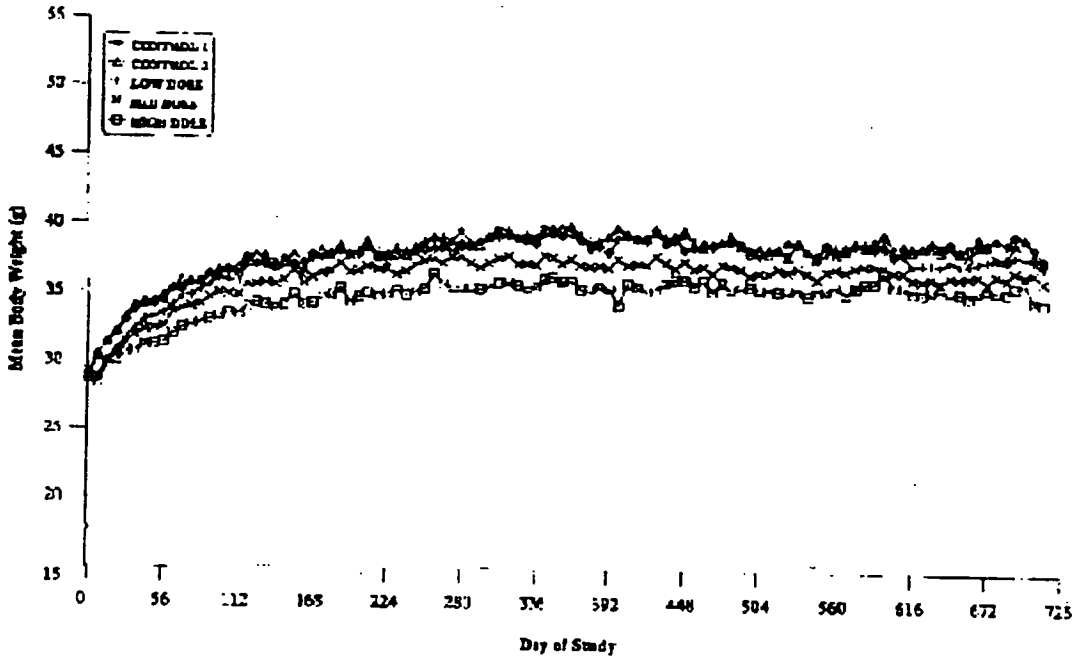
Mean Body Weight of Male Groups

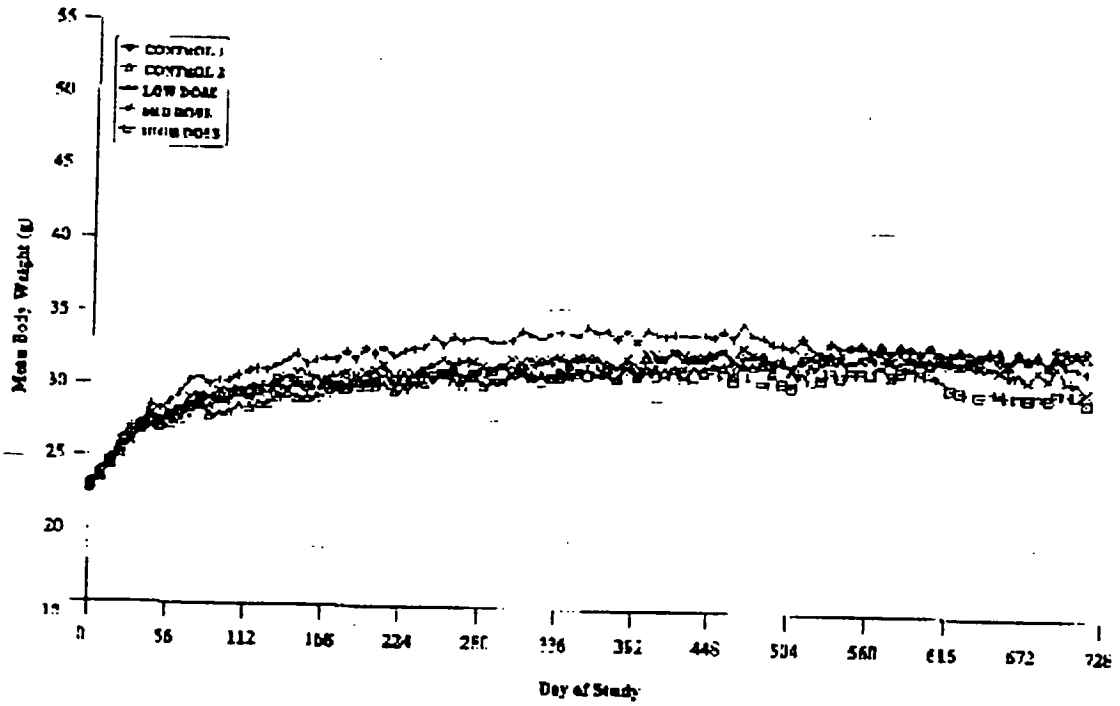
Study Number: 92114



In males, body weight was reduced (compared to CM) throughout the dosing period at the MD and HD, and for up to ≈Day 78 at the LD.

Mean Body Weight of Female Groups

Study Number: 92114



In females, body weight was increased (compared to CF) at the LD up to Day 512 and sporadically at the MD up to Day 337. Body weight was significantly reduced in MDF (compared to CF) only at the last measurement time (i.e., Day 715). At the HD, body weight was reduced (compared to CF) only from Day 617 on.

Food consumption: food consumption was reduced at all doses in males and females throughout the dosing period. By the end of the study, food consumption was reduced by 10, 13, and 13% at the LD, MD, and HD, respectively, in males and by 6, 8, and 5% at the LD, MD, and HD, respectively, (significantly only at the MD) in females.

Water consumption: in males, water consumption was reduced throughout the measurement period at all doses; by the end of the study, water intake was 14, 19, and 25% lower in LDM, MDM, and HDM, respectively, compared to CM. In females, water consumption was affected sporadically throughout the measurement period; however, the changes were not necessarily dose-related; by the end of the study, water intake in LDF, MDF, and HDF were 20, 7, and 28%, respectively, lower than in CF.

Ophthalmology: there were no apparent drug-related ophthalmology findings upon examination of the anterior segment of the eye.

Terminal studies

Gross pathology: no summary table was provided for gross pathology findings. Notable findings, according to the sponsor, are summarized in the following sponsor's table:

Incidence of noteworthy macroscopic observations on female animals (50 animals per group)

Organ	Macro.	Control 1	Control 2	Low	Mid	High
Ovary	Cyst	34	46	29	23	28
Uterus	Mass	12	8	4	2	0
	Enlarged	23	23	4	3	3
	Dilation	4	6	1	0	1
Pituitary	Mass	0	0	4	4	5
	Enlarged	1	0	1	7	19
Skin	Mass	3	4	12	9	35

Organ/tissue wts: there were no drug-related findings in females. In males, significant decreases in the absolute wt of brain (LD, HD), heart (LD, HD), kidney (all doses), and liver (HD) were noted. Relative wts of all of these but brain were also reduced (heart: 6% at HD; kidney: 8 and 10% at LD and HD; liver: 4% at HD).

Histopathology: selected non-neoplastic and neoplastic findings are presented in the attached tables (data are based on sponsor's summary tables).

Non-neoplastic findings, mouse carcinogenicity study

TISSUE	FINDING	MALES					FEMALES				
		C	C	LD	MD	HD	C	C	LD	MD	HD
adrenal	hyperplasia, spindle cells	13/50	17/48	13/50	21/50	26/50	46/49	43/49	38/50	36/50	39/49
	accessory cortical tissue	3/50	2/48	3/50	1/50	8/50	4/49	3/49	3/50	4/50	5/49
cervical node	plasmacytosis	1/45	1/46	0/47	0/47	0/43	0/44	0/45	1/45	1/42	4/48
Harderian gland	hypersecretion	0/50	0/50	2/49	2/50	2/49	1/50	1/50	4/50	4/50	7/50
lung	foam cell foci	1/50	3/49	4/50	7/50	6/50	1/50	5/50	5/50	3/50	11/50
ovaries	atrophy						8/50	5/50	10/49	24/50	35/49*
pituitary	hyperplasia, focal, pars distalis	2/47	1/48	0/46	0/46	1/47	1/47	1/45	6/48	14/50	13/48*
	hyperplasia, diffuse, pars distalis	0/47	0/48	0/46	0/46	0/47	0/47	0/45	0/48	11/50	16/48*
mammary gland	infiltration, mononuclear cells	0/49	0/49	0/48	0/49	0/50	1/50	0/47	1/49	0/49	6/50
	galactocoele	0/49	0/49	0/48	0/49	0/50	3/50	3/47	8/49	18/49	28/50*
	hyperplasia, lobular	1/49	0/49	0/48	0/49	0/50	0/50	1/47	22/49	23/49	25/50*
thyroid	infiltration, mononuclear cells	0/49	1/50	0/50	1/50	3/50	0/50	0/50	2/50	3/50	5/49
uterus	inactivity						0/50	0/50	2/50	8/50	12/49*

*p = 0.0001, trend test

Neoplastic findings, mouse carcinogenicity study

TISSUE	FINDING	MALES					FEMALES				
		C	C	LD	MD	HD	C	C	LD	MD	HD
Harderian gland	adenoma	4/50	3/50	5/49	6/50	9/49	1/50	0/50	5/50	2/50	0/50
lung	bronchiolar-alveolar adenoma	9/50	14/49	8/50	14/50	10/50	4/50	4/50	2/50	4/50	8/50*
pituitary gland	adenoma, pars distalis										
	fatal	0/47	0/48	0/46	0/46	0/47	0/50	0/45	0/48	0/50	1/48
	incidental	0/47	0/48	0/46	0/46	0/47	0/50	0/45	5/48	10/50	14/48**
	total	0/47	0/48	0/46	0/46	0/47	0/47	0/45	5/48	10/50	15/48**
	carcinoma, pars distalis	0/47	0/48	0/46	0/46	0/47	0/47	0/45	0/48	0/50	2/48
	adenoma, pars intermedia	0/47	1/48	0/46	1/46	0/47	0/47	0/45	1/48	1/50	2/48
mammary gland	adenocarcinoma										
	fatal	0/47	0/48	0/46	0/46	0/47	050	1/47	4/49	5/49	11/50**
	incidental	0/47	0/48	0/46	0/46	0/47	0/50	1/47	5/49	0/49	10/50**
	total	0/47	0/48	0/46	0/46	0/47	1/50	2/47	9/49	5/49	21/50**
	adenoacanthoma	0/47	0/48	0/46	0/46	0/47	0/50	0/47	0/49	0/49	1/50
	adenoma	0/47	0/48	0/46	0/46	0/47	0/50	0/47	0/49	2/49	0/50

*p = 0.04, trend test
**p = 0.0001, trend test

Pituitary adenomas in 10/10 HDF examined were prolactin-positive. In comparison, the sponsor noted that in pituitary adenomas examined in 3/3 CF in a previous study were not prolactin-positive.

Rat: dose selection for the rat carcinogenicity study was based on the results of 3-wk and 3-mo dietary studies in Fischer and Long-Evans rats, respectively. In the 3-wk study, ziprasidone was administered at doses of 0, 10, and 40 mg/kg for 2 wks and 0, 40, and 100 for an additional wk. A fourth dose-grp received 200 mg/kg for 3 days, but the dose was reduced to 100 mg/kg due to "poor toleration". Dose-limiting effects consisted of clinical signs (decreased activity, all doses) and reductions in body weight (compared to C); no drug-related deaths occurred. In males, mean body weight was reduced (compared to CM) by 9 and 16% at 10 and 40 mg/kg after 2 wks of dosing. In females, body weight was reduced by 6 and 8% at 10 and 40 mg/kg after 2 wks of dosing. The 3-mo study was conducted in the same strain used for the 2-yr study. In the 3-mo study, ziprasidone was administered at doses of 0, 5, 10, and 20 mg/kg. Doses were increased to final levels gradually over 1 mo; therefore, the MD and HD were tested only for 2 mo. The only drug-related effect observed was a reduction in body weight in males at the MD and HD (7 and 10%, respectively, as compared to CM). Body wt effects were minimal in females (4% below CF values at the HD).

The sponsor selected the HD for the 2-yr study on the basis of body wt effects in males and the fact that "The high dose of 12 mg/kg/day in feed in rats represents an approximate 5-fold multiple of the maximum projected clinical dose of 160 mg/day (80 mg b.i.d.) (i.e., 2.67 mg/kg/day for a 60 kg person)". At the time of the IND review for dose-selection, there were insufficient data to concur with the sponsor's proposed doses; however, it was this reviewer's opinion that the proposed HD was adequate for males, but too low for females.

2 year oncogenicity study with dietary administration in Long Evans rats (Study no. 92-720-21, Pfizer Central Research, study dates: 11/92-10/94, report date: 1/3/96, GLP, Vol. 1.23-1.26)

Animals: Long Evans rats (

initial body weight: 232 gm for males, 173 gm for females
housing: animals were housed singly.
n = 50/sex/grp + 16/sex/grp (LD, MD, HD grps only) for TK analysis

Drug: ziprasidone hydrochloride (CP-88,059-1; lot no. 20480-236-1MS, 23638-297-1F, 23638-95-1F)

storage: the drug/lactose "preblend" was stored at rm temperature, protected from light. The drug-diet admixtures were stored at 5° C, protected from light. The lengths of storage was not specified.

stability: the stability of drug in diet was tested at 30-40° C with light challenge. According to the sponsor, the drug mixture was stable for 1 wk at rm temperature and for 4 wks at 5° C. Open container challenge didn't affect the stability; however, light challenge resulted in loss at low dose levels. Stability data were not provided.

vehicle: ground rodent diet and lactose diluent; controls received lactose comparable to the HD grps.

formulation analysis: aliquots from each drug-diet admixture were assayed for achieved drug concentration. According to the sponsor, all samples met the accepted range of potency limits of 85-115% except for the LD diet during Wks 69-72 which were 78-86% of intended. Again, according to the sponsor, this result was due to "assay difficulties".

doses: 0, 0, 2, 6, and 12 mg/kg. According to the sponsor, the concentration of drug in

diet was adjusted "...according to projected values for body weight and food consumption". Analysis of the diet indicated that achieved doses were 94-116%, 87-122%, and 79-112% of intended at the LD, MD, and HD, respectively. Dose for all treated grps was initially 2 mg/kg/day. After 2 wks, MD and HD were increased to 6 mg/kg/day, and after 4 wks, the HD was increased to 12 mg/kg/day.

route: p.o. (dietary)

Observations

Clinical signs: all animals were observed daily. Clinical signs were examined biweekly throughout the dosing period, and palpable masses were checked once a month beginning at 6 mos.

Body weight: body weights were recorded weekly beginning 1 wk prior to the start of dosing.

Food consumption: food consumption was measured weekly up to 6 mo and monthly from 6 mo on. According to the sponsor, "Values associated with suspected food jar scratch-outs were not recorded".

Ophthalmology: ophthalmology examinations were conducted once prior to the start of dosing, and on Days 366, 554, and 715 of dosing. Mydriasis was induced prior to examination by application of 1.0% tropicamide.

Hematology: blood samples were collected by orbital sinus puncture from 10/sex/grp on Days 161, 357, 561, and "...from all survivors on Days 722-724 (males) and 726-730 (females)" for analysis of the following parameters: rbc, hgb, MCV, wbc (total, differential), platelet ct, MCH, MCHC.

Clinical chemistry: blood samples were collected by orbital sinus puncture from 10/sex/grp on Days 161, 357, 561, and "...from all survivors on Days 722-724 (males) and 726-730 (females)" for analysis of the following parameters: Na, K, Ca, Cl, BUN, creatinine, ALT, AST, SDH, total protein, albumin, 5'-nucleotidase; globulin, A/G were calculated.

Urinalysis: urine samples were collected over an ≈5-hr period "...the day before each bleeding interval" for analysis of the following parameters: volume, specific gravity, pH, protein, blood, glucose, urobilinogen, bilirubin, ketones, color.

TK: blood samples were collected from the vena cava in satellite animals on Days 197-198 of dosing. Samples were collected at ≈6, 12, 18, and 24 hrs from the beginning of the dark phase. Ziprasidone levels were determined in plasma using . : the LLOQ was . ng/mL.

Terminal studies

Gross pathology: a complete necropsy was conducted in all survivors (Days 722-730).

Organ/tissue wts: wts of the following organs/tissue were recorded: kidney, liver, brain, heart, adrenals (combined), and testes (combined).

Histopathology: microscopic analysis of tissues was conducted in all animals. Sections were read by two independent pathologists.

Conventions followed in this study: the following conventions were noted by the sponsor:

(1) "The coexistence of two or more malignant neoplasms of a single cell type in a single tissue, e.g., HEPATOCELLULAR CARCINOMA, is recorded as a single neoplasm."

(2) "The coexistence of a malignant and a benign neoplasm of the same cell type in a single tissue, e.g., HEPATOCELLULAR CARCINOMA and HEPATOCELLULAR ADENOMA, is recorded as two separate neoplasms."

(3) "...Malignant neoplasms [other than lymphomas] with metastases are recorded under the tissue determined to be the primary site of the neoplasm. Metastases are recorded under the tissue in which found and assigned the relationship to death code "Multicentric". If the primary site can not be determined the neoplasm is recorded as a metastasis under the tissue in which found."

(4) Neoplasms originating in the reticuloendothelial system were designated "Lymphoreticular", and the tissue of origin was not determined. "...the diagnosis and relationship to death code assigned under 'Lymphoreticular' are not recorded elsewhere". Related macroscopic findings were designated "HEMO/LYMPHO/RETIC NEOPLASM, with the relationship to death code listed as "multicentric". Lymphomas were statistically analyzed using the data listed under "lymphoreticular", but not "HEMO/LYMPHO/RETIC NEOPLASM".

(5) The death code categories, used only for neoplasms, were as follows: (1) definitely incidental, (2) probably incidental, (3) probably fatal, (4) definitely fatal, and (5) multicentric.

(6) The "C/D" designation refers to "cause of death" and were determined taking into account 11 organ systems.

Statistics: final body weight, weight gain, clinical pathology parameters, and organ/tissue wt data were analyzed statistically, using the appropriate test (e.g., Dunnetts multiple comparison test, Cochran-Cox modified t-test). Survival data were analyzed using the log rank test (SAS PROC LIFETEST). Tumor incidences (for those occurring in ≥5 animals) were analyzed using the Cochran-Armitage Trend test, and were mortality-adjusted. Significance was determined as per Peto *et al*, 1980.

The two control grps (per sex) were combined for analysis.

Results

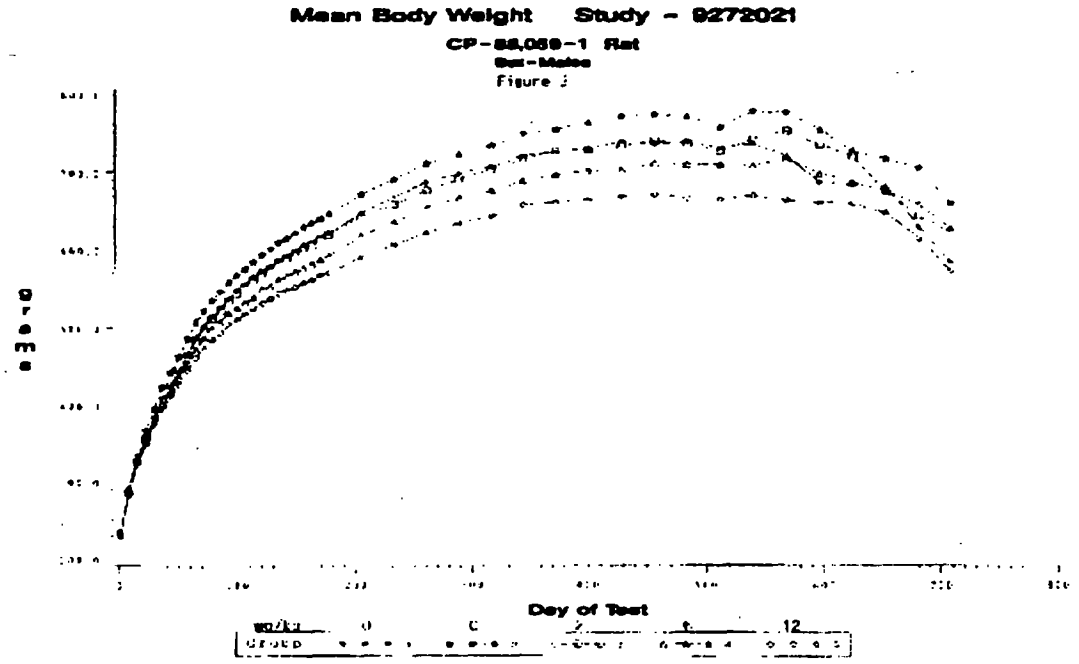
Mortality: there were no drug-related increases in mortality. There was a tendency for survival rate to be higher in HD grps (25-30% vs 14-23% in C grps).

Clinical signs: according to the sponsor, there were no drug-related clinical signs.

Ophthalmology: there were no apparent drug-related findings (C and HD grps only were examined, 25/sex/grp).

Body weight: body weights changes were provided in the sponsor's tables (below). In males, body weight was reduced (compared to CM) at the MD and HD (maximum of 5 and 9-10%, respectively) from Day 113-176 and Day 57-568, respectively. However, by the end of the dosing period, body weights were fairly similar among grps, due, in part, to mean body weight loss in the reference C grp (designated C1). Overall mean body weight gain was similar among grps.

In females, body weight was lower (compared to CF) in dose grps throughout the dosing period. Up to Day 372, however, the effect ranged from 4-7% and was not dose-related and not always statistically significant. By Day 400, body weight in HDF was significantly lower than in CF and remained so throughout the rest of the dosing period (maximum effect, 19% on Day 680). During this period, body weight tended to be lower in LDF and MDF as well, achieving statistical significance at the LD (maximum effect, 15% on Day 680). Overall mean body weight gain was significantly reduced only at the HD (22%).



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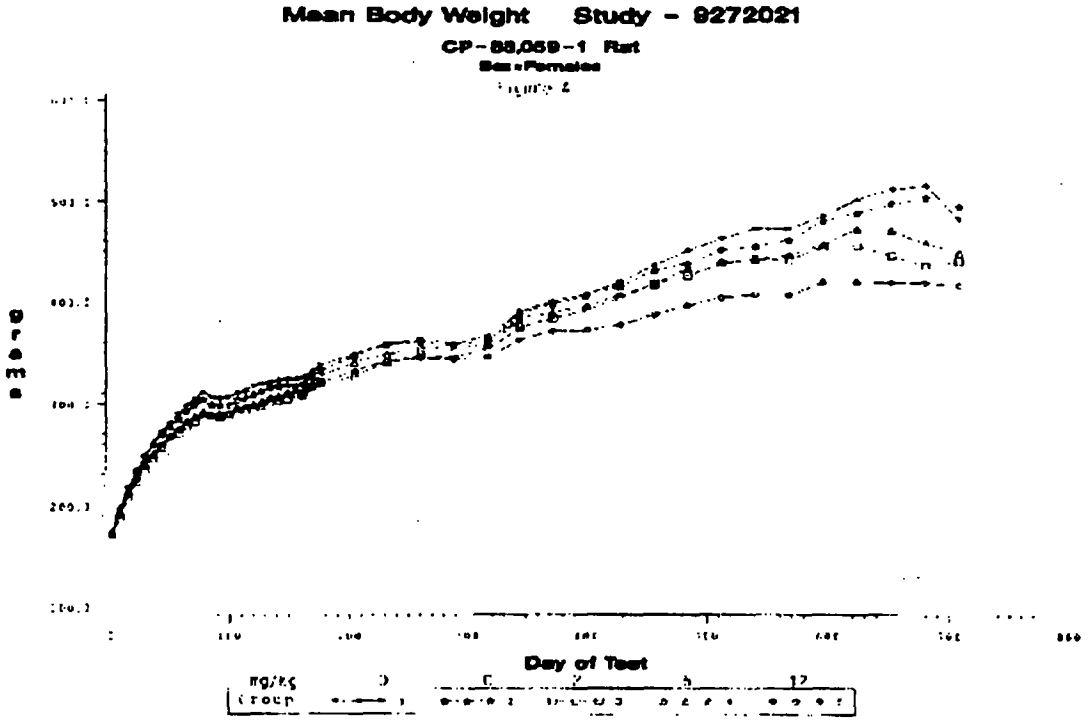


Food consumption: in males, food consumption was lower throughout most of the dosing period at the HD and during portions of the dosing period (particularly during Days 148-344) at the MD. From Day 596 on, food consumption was slightly elevated in HDM (≤22%) when compared to the reference CM (but fairly similar to the second CM grp).

In females, food consumption was lower at the HD throughout the dosing period, and during the first 78 days of dosing at the lower doses.

Hematology: there were no apparent drug-related changes. Wbc counts were elevated sporadically in both C and HD grps in males.

Clinical chemistry: there were no marked drug-related effects. Total protein was and albumin were reduced at all doses in males on Day 722 (total protein: 7, 9, and 10% at the LD, MD, and HD, respectively; albumin: 10-11%). Sodium was slightly (but significantly) reduced in HDM and HDF (2%) on Day 722.

TK: plasma levels of CP-88,059 are summarized in the following table (C_{max} data are expressed as mean ± SD):

DOSE (mg/kg)	MALES		FEMALES	
	C_{max} (ng/mL)	$AUC_{(0\text{-}24\ hr)}$ (ng•hr/mL)	C_{max} (ng/mL)	$AUC_{(0\text{-}24\ hr)}$ (ng•hr/mL)
2	21.2 ± 5.1	386	46.3 ± 14.2	758
6	70.5 ± 5.9	1274	132.7 ± 21.0	2162
12	111.5 ± 22.0	2040	250.8 ± 102.3	4193

*highest plasma level recorded, at 6 hr after start of dark cycle

The sponsor did not provide estimates of variability for the AUC data, nor individual AUC data.

Plasma exposure (C_{max} , AUC) was ≈2-fold higher in females than in males at all doses.

Terminal studies

Gross pathology: there were no clear drug-related findings.

Organ/tissue wts: the only effects were decreases in wts of several organs (i.e., liver, heart, and adrenal) in HDF due to decreases in body weight; relative wts of these organs were similar to or slightly higher than in CF.

Histopathology: findings are summarized in the attached table. The only significant findings were an increase in ß-keratoacanthoma (incidental) in HDM and an increase in mammary gland fibroadenoma in HDF. [According to the sponsor, when adjusted for multiple pair-wise comparisons (using Bonferonni), the finding in HDM was no longer statistically significant.] The finding in females, however, would not be statistically significant using the recommended p-values for common tumors.

The eye mineralization was characterized as "slight to mild focal to multifocal corneal mineralization" by the sponsor, but was not considered a drug-related finding.

histopathology findings (non-neoplastic, neoplastic), 2-yr oral carcinogenicity study in rats

TISSUE	FINDING	MALES					FEMALES				
		C1	C2	LD	MD	HD	C1	C2	LD	MD	HD
pancreas	acinar atrophy	8/48	8/50	11/49	14/49	11/49	4/50	5/48	13/49	13/48	10/50
	ß-islet cell adenoma	1/48	4/50	4/49	1/49	6/49	5/50	2/48	2/49	2/48	0/50
ovaries	ß-thecal/granulosa cell tumor						0/50	0/49	0/50	0/49	2/50
uterus	polyps						0/50	1/49	2/50	1/49	4/50
skin	ß-kerato-acanthoma	0/49	1/49	1/50	0/49	4/49*	1/49	0/49	0/50	0/49	0/50
mammary gland	adenocarcinoma	0/49	0/49	0/50	0/49	2/49	4/49	1/49	0/50	0/49	3/50
	fibroadenoma	0/49	0/49	1/50	0/49	0/49	16/49	15/49	14/50	17/49	20/50**
eye	mineralization	7/48	11/49	12/48	14/48	19/50	2/49	8/47	10/48	4/48	8/50

*p = 0.0079 using Cochran-Armitage test for linear trend; p = 0.1106 using Bonferroni adjustment for multiple tests. Analyzing the HD against each C grp separately, p = 0.0091 and 0.0234 for C1 and C2, respectively.

**p = 0.0490 using Cochran-Armitage test for linear trend, CF vs C2 only.

The following studies have been previously reviewed: Segment I and Segment II studies in Sprague-Dawley rats, Segment II study in New Zealand White rabbit. Summaries of these studies are provided.

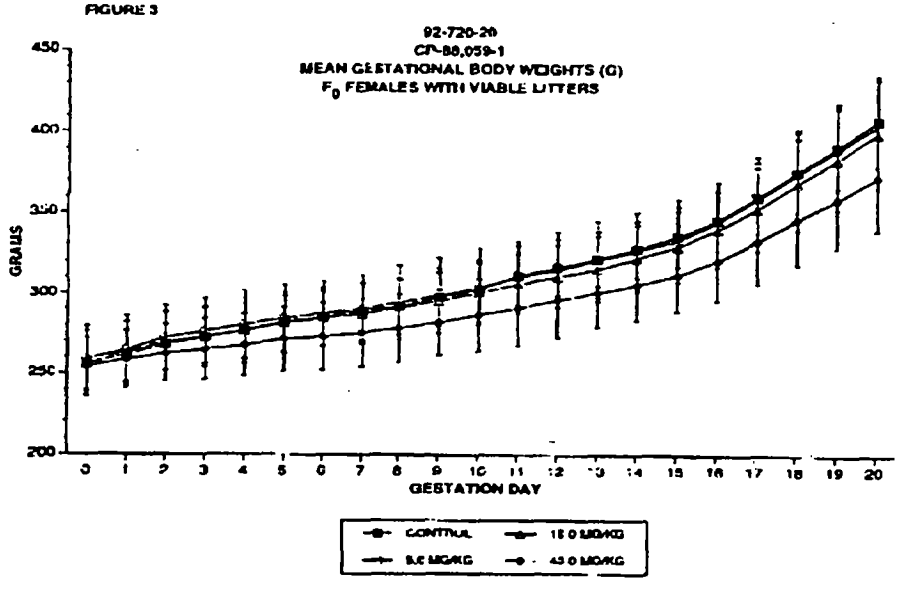
Segment I: mating and fertility

This study was previously reviewed (P/T review, 3/22/94, L.M. Freed, Ph.D.); the methods and results are summarized here (summary data are provided).

A Segment I (mating and fertility) study was conducted in Sprague-Dawley rats (n = 20/grp for males, 40/grp for females) at doses of 0, 5, 10, and 40 mg/kg (gavage). Females were dosed starting 15 days prior to mating and continuing throughout the lactation period (to Day 21 postpartum). Males were dosed for 72 days prior to and during the mating period; each male was mated with 2 females. A Cesarean section was performed on approximately one-half of the females and fetuses were examined for visceral (Wilson's technique; 1/2 of fetuses) and skeletal findings (Alizarin red; 1/2 of fetuses). The remaining dams were allowed to deliver naturally. Live pups were culled to 4/sex/litter (where possible) on Day 4 and the selected pups were assessed for achievement of developmental/behavioral landmarks during lactation. At weaning, 2/sex/litter continued to be followed for physical/behavioral development and at Days 90-117 postpartum were mated to assess reproductive capability.

There were no drug-related deaths during the study. Clinical signs consisted primarily of sedation (dose-dependent), which was noted throughout the dosing period, and chromodacryorrhea (MDM, HD animals). Body weight was reduced in a dose-related manner in males, with the effect being significant only at the MD and HD (10-18%). In females, body weight during gestation was significantly lower (as compared to CF) at the HD (maximum of 9% on Day 20 of gestation) (cf. sponsor's Fig. 3; **note that the ordinate does not start at zero**).

FIGURE 3

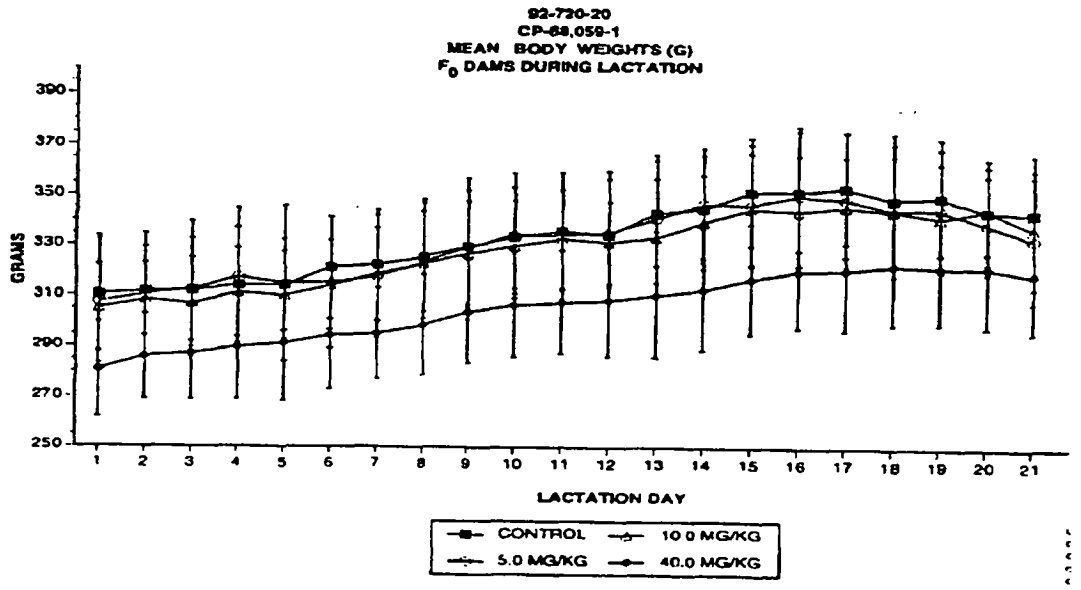


Body weight gain was reduced primarily at the HD and primarily during the first wk of gestation (36% vs 23 and 16% at Wk 2 and Wk 3, respectively). During Wk 2, body wt gain was significantly reduced at all doses (11, 13, and 23% in LDF, MDF, and HDF, respectively); however, mean body wts on Day 14 were within 0-2% of CF at the LD and MD (327.8 ± 17.6,

326.6 ± 23.4, 322.1 ± 20.4 in CF, LDF, and MDF, respectively). **[It should be noted that the sponsor's summaries of the body wt data (tables, figs) during gestation included only those dams with viable pups and not all pregnant dams. It should also be noted that there was a discrepancy between the no. of dams per grp listed in the summary table and the no. of pregnant dams with viable pups listed in the individual data tables. According to the individual tables, there were 33, 36, 35, and 33 CF, LDF, MDF, and HDF, respectively, whereas the summary table listed "n'" as 32, 31, 34, and 32, respectively. Even with missing body wt data taken into account, the "n's" do not agree.]**

Changes in body wt during lactation are illustrated in the following sponsor's Fig 5 (**note that the ordinate does not start at zero**). Body wt was reduced BY 7-10% at the HD (compared to CF) throughout the lactation period; however, the differences between these two grps achieved statistically significant only sporadically.

FIGURE 5



Body wt gain during lactation was fairly similar among grps and was significantly higher in HDF during the last wk of lactation (-8.5 ± 11.5, -13.5 ± 14.5, -7.7 ± 18.3, and 2.1 ± 14.5 g in CF, LDF, MDF, and HDF, respectively).

In males, food intake was reduced at all doses during the premating period (8-17%). In females, food intake was reduced only in HDF and only during Wks 1 and 2. During lactation, food intake was reduced by 11-19% in HDF. On fertility parameters (taking all dams into account), the following effects were observed: (1) an increase in the cohabitation interval (i.e., time to sperm-positive vaginal smear). The median time was 3, 4, 3-4, and 5-7 for CF, LDF, MDF, and HDF, respectively. The sponsor considered only HDF affected. The median interval was 1-4 in CF, LDF, and MDF, and 5-7 days in HDF; at the HD, a sperm-positive vaginal smear was not obtained in 10 females until ≥Day 10. The number of females with sperm-positive vaginal smears at Day 10 or later was 3, 3, 6, and 10 CF, LDF, MDF, and HDF, respectively. (2) an increase in the no. of days during the premating period with no sign of estrus in all dose grps (1/40 CF, 7/40 LDF, 12/40 MDF, and 13/40 HDF). [The sponsor noted that "...days on which estrus was observed were recorded..."; however, "There was a large individual variation between animals within each dose group and because all stages of the estrous cycle were not

recorded, clear patterns did not emerge after one week of examination."] No drug-related effects were noted on the number of sperm-positive vaginal smears, pregnant females, total copulation rate (98-100%), or total pregnancy rate (85-90%).

The summary data on reproductive parameters for dams delivered by Cesarean section were provided in the following sponsor's table:

MEAN REPRODUCTIVE DATA IN F_0 FEMALES ON GESTATIONAL DAY 20

Dose Group	No. of Litters	Corpora Lutea	Implantation Sites	No. of Fetuses		No. of Resorptions	Mean Fetal Body Wt. (G)		Ratio M/F Pups
				Viable	Dead		male	female	
Control	16								1.00
Mean		16.2	14.6	13.9	0.00	0.69	3.92	3.72	
s.d.		2.8	3.6	3.3		0.70	0.29	0.30	
5.0 mg/kg	16								1.11
Mean		17.5	15.7	14.9	0.06	0.75	3.78	3.56	
s.d.		2.0	2.7	2.6	0.25	1.06	0.19	0.20	
10.0 mg/kg	18								1.02
Mean		17.3	15.2	14.2	0.00	1.00	3.64 ++	3.47 +	
s.d.		2.6	3.7	2.9		1.03	0.35	0.38	
40.0 mg/kg	15								0.80
Mean		15.6	13.4	12.4	0.00	1.00	3.58 ++	3.36 ++	
s.d.		3.0	3.7	4.3		1.93	0.21	0.22	

+ $p < 0.05$ ++ $p < 0.01$

The only significant drug-related finding was a decrease in mean fetal (M/F) body wt in MDF (7%) and HDF (9%); the 4% decrease in mean fetal body wt for LDF was not statistically significant. Although not statistically significant, the following were also noted: (1) the no. of corpora lutea, implantation sites, and of viable fetuses were slightly reduced at the HD (4-11%), (2) the no. of resorptions was increased (45%) at the MD and HD, and (3) the ratio of M/F pups was lower (20%) at the HD.

Examination of fetuses indicated no apparent drug-related external or visceral findings. The sponsor provided a summary of selected skeletal findings and expressed the summary data in terms of an "ossification index" (mean ± SD) or number of affected fetuses. The data expressed as "ossification index" could not be compared to the individual data. In addition, the sponsor stated that "Only fetuses without skeletal variations in bones listed...were included in the ossification index". [The sponsor has been asked to provide summaries expressed as affected fetal and litter incidences.] The following table summarizes the drug-related skeletal findings as reported by the sponsor in Table 9:

FINDING	C	LD	MD	HD
Sternebrae ossification index # fetuses with fully ossified sternum	 4.83 ± 0.71 33	 4.58 ± 0.48 15	 4.35 ± 1.20 21	 4.19 ± 0.50** 4
Metacarpals ossification index, R ossification index, L	 3.61 ± 0.37 3.57 ± 0.37	 3.44 ± 0.38 3.47 ± 0.40	 3.21 ± 0.42** 3.21 ± 0.39**	 3.27 ± 0.39** 3.26 ± 0.43**
Sacral/caudal vertebrae (# of fetuses) marked	15/115	18/123	36/132	22/98
Hyoid, absent ossification (# of fetuses)	10/113	24/118	28/129	21/98
Pubis, absent ossification (# of fetuses)	0/115	0/123	4/133	1/98

**p<0.01

The reproductive parameters in dams allowed to deliver naturally were summarized in the following sponsor's table:

MEAN F_1 PUP VIABILITY POSTNATAL DAY 1 TO WEANING

Dose	No. of Implantations	No. Born Total	No. Born Alive	% Born Dead	No. (%) Surviving to: Day 1	Day 4*	Day 7**	Day 10	Day 14	Day 21
Control	17.1 ± 3.9	15.6 ± 2.4	15.5 ± 2.4	1.1	15.5(100.0)	15.1(97.5)	7.9(100.0)	7.9(100.0)	7.9(100.0)	7.9(100.0)
5 mg/kg	16.6 ± 2.5	15.2 ± 2.4	15.0 ± 2.7	1.8	14.9(99.7)	14.5(95.7)	8.0(100.0)	8.0(100.0)	8.0(100.0)	8.0(100.0)
10 mg/kg	16.4 ± 2.5	15.2 ± 3.0	13.3 ± 4.6	11.0 +	13.7(100.0)	13.4(98.6)	7.4(100.0)	7.3(98.4)	7.2(96.1)	7.1(96.1)
40 mg/kg	15.7 ± 1.9++	14.5 ± 2.0	13.3	7.1++	7.9++	13.8(96.3)	10.9(82.5)	7.2(99.3)	7.2(99.3)	7.2(99.3)

* calculations for Day 4 are before culling and are based on number of pups born alive
** calculations for Days 7, 10, 14 and 21 are based on litters after culling on Day 4
a) the mean percentage for the 10 mg/kg dose group was high because of one litter with greater than 60% dead. The nonparametric trend test performed on this variable found the group difference not to be significantly different.
++ p < 0.01

There were no apparent drug-related effects on the length of gestation. However, decreases in implantation sites were noted in HDF and the number of live pups (per litter) was reduced at both the MD and HD. As noted in the above table, the majority of pups in one MD litter were stillborn which did contribute to the overall higher percentage of dead pups at the MD. However, the percentage dead in the affected MD litter was 59% (i.e., 10/17), not "greater than 60%". In addition, the no. of litters in which at least one pup was stillborn was increased at the MD and HD (3/17, 3/20, 7/17, and 10/18 affected litters in CF, LDF, MDF, and HDF, respectively). There was also a decrease in survival rate on Day 4 at the HD, although the effect was not statistically significant. [Individual survival data were not provided.]

In F_1 pups, body wt was reduced throughout the lactation period in HD male and female pups (9-14%) [birth wts, i.e., Day 0, were not provided]. After weaning, body wt was still reduced in HD pups during the first wk after weaning; however, over the next 3 wks, body wt tended to normalize (more rapidly in male pups) and by Day 49 postpartum, there were no significant differences among grps.

In terms of developmental milestones, no drug-related effects were noted on pinna

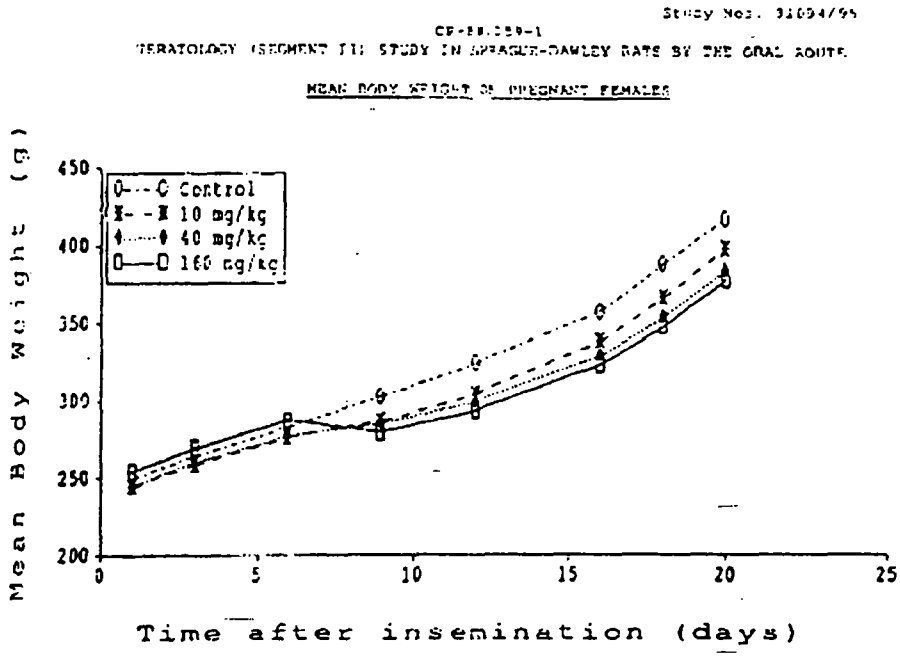
detachment, surface righting, eye opening, visual cliff avoidance, or motor activity. Delays (expressed as the % of litters meeting criterion on test days) in the following were noted: (1) negative geotaxis in HDM, (2) eruption of incisors in HDF, (3) grip strength in MDM and HDM, and (4) air righting at all doses in males. By the end of testing, the only effect still evident was a delay in air right in HDM. Postweaning, there were no significant drug-related effects on locomotor activity, vaginal opening, and preputial separation; however, horizontal counts tended to be increased in females in a dose-related manner (2-24%). Reproductive performance was not adversely affected in F_1 animals.

Segment II studies: embryo/fetal toxicity

Segment II studies in Sprague-Dawley rat (Study no. 91094-95) and New Zealand White rabbit (Study no. 91096/97) have previously been reviewed (P/T review, 3/22/94, L.M. Freed, Ph.D.); the methods and results are summarized here (summary data are provided).

Rat. Ziprasidone was administered orally by gavage (n = 20/grp) at doses of 0, 10, 40, and 160 mg/kg; an additional grp of rats was dosed at 160 mg/kg (n = 5) for collection of TK data. Animals were dosed from Day 6 through Day 17 of gestation (i.e., postinsemination). Dams were sacrificed on Day 20 of gestation and fetuses were examined for external, visceral (Wilson's technique), and skeletal (Alizarin red) findings. TK data were collected on Day 17 (1, 3, and 5 hr postdosing).

There were no unscheduled deaths during the study. Clinical signs, consisting of ptosis, prostration, piloerection, and dyspnea were observed in all MD and HD animals. Body weight was reduced throughout the dosing period at all doses (3, 7, and 10% in LDF, MDF, and HDF, compared to CF; data illustrated in following sponsor's fig.; **note that the ordinate does not start at zero.**)



There were no apparent gross pathology findings in the dams. In terms of litter parameters,