there were no drug-related effects on no. of corpora lutea, implantation sites, viable fetuses, dead implants, or male/female ratio. Fetal body wt was lower in all dosed grps (male and female); however, the effect was statistically significant at the MD and HD in males, and at the HD in females (data provided in the following sponsor's Table 4).

MALES

	CONTROL	10 MG/KG	60 MG/KG	160 MG/KG
MEAN	3.91	3.78	3.71	3.57
ADJ. MEAN	3.93	3.78	3.74	3.59
ST.ERROR	0.06	0.06	0.06	0.06
N OF LITTERS	20	18	19	18
SIGNIFICANCE		N.S.	P<0.05	P<0.001

FEMALES

	CONTROL	10 MG/KG	60 MG/KG	160 MG/KG
MEAN	3.71	3.56	3.55	3.41
ADJ. MEAN	3.69	3.57	3.57	3.42
ST.ERROR	0.06	0.06	0.06	0.06
N OF LITTERS	20	18	19	18
SIGNIFICANCE		N.S.	N.S.	P<0.01

There were no clear drug-related external findings; flexed paws were detected in 1 LD fetus and 4 HD fetuses (1 litter). Hematomas were observed in 2-4 fetuses in each grps, including controls. There were also no drug-related visceral findings. The sponsor provided summaries only of selected skeletal findings, i.e., variations in ribs and degree of ossification of selected bones. The sponsor has been asked to provide summaries of all skeletal findings. Based on the summary table provided in the original report, the following appear to be drug-related (data expressed only as # of affected fetuses):

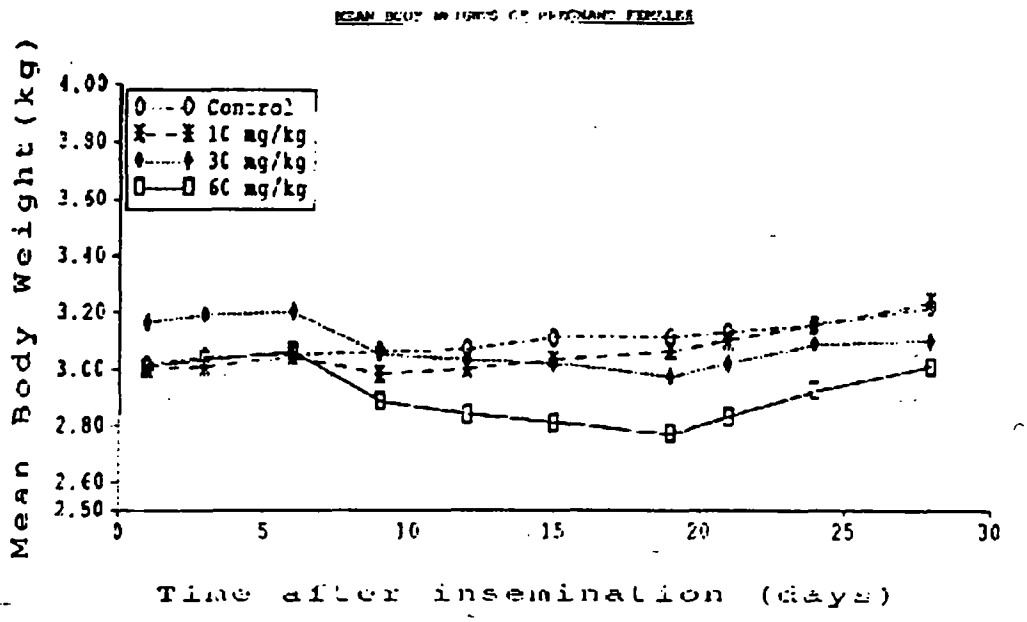
FINDING	C	LD	MD	HD
Sacral/caudal vertebrae slight delay ossification	22/135	26/123	35/109	32/108
5th metacarpus				
absent ossification, R	51/157	47/149	56/145	72/140
absent ossification, L	56/157	49/149	58/145	74/140

PK data were collected in satellite animals (5/grp) on Day 17 (the last day of dosing). The concentration of ziprasidone in maternal plasma, amniotic fluid, and fetal homogenates were quantitated. Mean levels (± SD) were 3.66 ± 1.31 (1-hr value), 0.41 ± 0.15, and 1.31 ± 0.08 µg/mL or g wet wt, respectively.

Rabbit. Ziprasidone was administered to New Zealand White rabbits (n = 20/grp, 4 rabbits for satellite-TK study) at doses of 0, 10, 30, and 60 mg/kg by gavage from Day 6 through Day 18 post-insemination (Study No. 91096/97). Dams were sacrificed on Day 28 of gestation and fetuses were examined for visceral [using a "...modification of the original Staples and Schnell method..."; quote from the sponsor) and skeletal (Alizarin red stain) findings. Unfortunately, one-half of fetuses were examined for visceral findings and the remaining fetuses were examined for skeletal findings. In rabbits, the size of the fetuses allows the examination of each fetus for both visceral and skeletal findings. Satellite-TK animals were sacrificed on Day 18 and blood, amniotic fluid, and fetal tissue were collected for analysis of ziprasidone levels.

There were 2 unscheduled deaths during the study: 1/20 LDF, 2/20 HDF. The death at the LD was not clearly drug-related; the animal was found trapped in the cage with hindlimb injuries and was sacrificed on Day 19 of gestation. Both HD deaths (Days 27 and 18) were spontaneous, following a period of reduced food intake and diarrhea. The only clearly drug-related clinical sign, as described by the sponsor, was blood in the cage, noted in 1/20 MDF

and 3/20 HDF. Prostration was noted only in 1 HDF. Body weight loss was noted in MDF and HDF during the dosing period, and in LDF only during the first few days of dosing (i.e., on Day 9). The maximum effect occurred on Day 19 at which body weight was 4 and 11% lower, respectively, than in CF. The data are illustrated in the following sponsor's Fig.1 (**note that the ordinate does not start at zero**):



Food intake was not quantitatively assessed. According to the sponsor, the incidences of "reduced intake" or "no intake" was slightly higher in dosed grps during the dosing period in pregnant dams (10/19, 13/18, 16/17, and 16/16 in CF, LDF, MDF, and HDF, respectively). After the dosing period, the incidence of reduced/absent food intake was still higher in MDF and HDF. However, the categorical nature of the data make them difficult to interpret. There were no clearly drug-related gross pathology findings in dams at necropsy. The data on litter parameters are summarized in the following sponsor's Table 1:

	CONTROL	10 MG/KG*	30 MG/KG	60 MG/KG
REPRODUCTIVE VARIABLES FOR SACRIFICED FEMALES				
PREGNANCY RATE (%)	19/ 20 (95)	17/ 19 (89)	17/ 20 (85)	14/ 18 (78)
VIABLE LITTERS ON DAY 20 (%)	16/ 19 (84)	17/ 17 (100)	12/ 17 (71)	8/ 14 (57)
CORPORA LUTEA MEAN±S.D.	8.4± 1.90	7.8± 2.56	8.3± 1.71	7.9± 2.30
IMPLANTATION SITES MEAN±S.D.	6.1± 2.93	5.5± 2.61	6.8± 2.63	6.8± 3.20
NO FOETUSES MEAN±S.D.	5.2± 2.59	5.3± 2.87	5.5± 2.61	5.3± 2.64
IMPLANTATION RATE (%)	97/135(71.9)	88/125(70.4)	81/111(73.0)	54/ 63(85.7)
EMBRYOMORTALITY RATE (%)	14/ 97(14.4)	3/ 88(3.4)	15/ 81(18.5)	7/ 54(13.0)
FOETAL DEVELOPMENT				
MEAN FOET.WEIGHTS	34.66± 5.93	35.81± 3.76	30.94± 6.98	28.55± 6.47
MEAN BODY WEIGHT GAINS OR LOSSES OF PREGNANT FEMALES (grams)				
NO OF PREGNANT FEMALES	16	16	12	8
FROM DAY 1 TO DAY 20	201.2	246.3	-69.1	-3.0
FROM DAY 1 TO DAY 5	37.7	39.3	32.3	45.9
FROM DAY 6 TO DAY 19	59.8	24.3	-231.4	-294.6
FROM DAY 19 TO DAY 28	103.8	182.8	130.1	245.8

* F60% not taken into account, except for pregnancy rate and viable litters on day 20 p.c.

Due to a combination of mortality in dams, reduced pregnancy rate, and/or reduction in viable litters at the MD and HD, there were an insufficient number of litters/fetuses available for examination at these doses. Also, 1 MDF (Day 28) and 2 HDF (Days 17 and 22) aborted their entire litter. [Note: the sponsor was asked to repeat this study.]

Based on this reduced number, however, certain observations were made. The no. of litters with at least 1 viable fetus was reduced in MDF and HDF. Unfortunately, data from dams that had no viable fetuses were not included in means for implantation or embryomortality (i.e., postimplantation loss) rates. [Note: the sponsor should be asked to re-calculate the means using data from all pregnant dams; a quick calculation of postimplantation loss with all data included resulted in %'s of 26 and 37%.] Mean fetal body wt was reduced in MDF and HDF (11 and 18%, respectively), however, the effect was statistically significant only at the HD. The inadequate no. of litters/fetuses in the MD and HD was compounded by the examination of only one-half of fetuses for visceral and the remaining fetuses for skeletal. This resulted in the following no. of fetuses examined:

visceral examination: 42, 48, 33, and 22 pups in C, LD, MD, and HD grps.
skeletal examination: 41, 45, 33, and 25 pups in C, LD, MD, and HD grps.

The no. per grp was inadequate for all grps, including controls. Based on these reduced numbers, no drug-related visceral or skeletal findings were evident. For the skeletal data, only selected findings were reported, and the data were expressed only as the number of affected fetuses. The sponsor has been asked to provide summaries of all skeletal findings, expressed as no. (%) of affected fetuses and litters.

The PK/TK data were minimal; only 4 dams were evaluated and of these 4, 1 was not pregnant and 1 produced no viable fetuses. The plasma C_{max} (based only on 1, 3, and 5 hr values) was 1.24 µg/mL (range: _/mL). In the two dams that were pregnant and had viable fetuses, plasma, amniotic fluid, and fetal tissue levels were 0.77-1.42, 0.06-0.07, and 0.25-0.26 µg/mL or µg/g wet wt, respectively.

The following studies have not been previously reviewed; reviews follow.

1. **Preliminary Segment I fertility and reproduction study in Sprague-Dawley rats** (Study no. 92-720-17, study dates: 3/9/92-5/21/92, GLP, Vol. 1.27).

Methods: CP-88,059-1 (lot no. 20,480-236-1MS; vehicle: 0.5% MC) was administered to Sprague-Dawley rats (10/sex/grp) at doses of 0, 0, 10, 40, and 160 mg/kg p.o. (by gavage). Males received daily doses from 4 wks prior to mating through the mating period; females received daily doses from 29 days prior to mating, and through mating, gestation, and lactation (up to Day 9-11 postpartum). The mating scheme used is summarized in the following sponsor's table:

Males	Seq. #	Mated To	Females	Seq. #
Control	1-10	X	Control	21-30
Control X	11-20	X	160 mg/kg X	31-40
10 mg/kg	41-50	X	10 mg/kg	51-60
40 mg/kg	61-70	X	40 mg/kg	71-80
160 mg/kg	81-90	X	160 mg/kg	101-110
160 mg/kg X	91-100	X	Control X	111-120

This scheme, as noted, included 2 cross-mating grps. The presumed first day of pregnancy was designated as Day 0. Observations in treated animals included: body wts, litter parameters (live/dead pups), gross pathology (Day 10 postpartum). Non-pregnant F_0 females were sacrificed on Day 24 of intended gestation, and necropsied.

All live F_1 pups were raised to Day 4 postpartum. On Day 4, litters were culled to 4/sex/litter and these pups were followed up to Day 10 postpartum. Observations in F_1 animals were as follows: body wt, survival, gross pathology (Day 10 postpartum).

F_0 males were sacrificed on Day 45 and necropsied.

Results: there were 2 unscheduled deaths during the study: 1 CF (dystocia, Day 40), 1 HDF (gavage accident, Day 17). The primary drug-related clinical sign was sedation, with the severity being dose-related. Other clinical signs included chromodacryorrhea (MD, HD) and poor grooming (HDM). Prior to mating, body wt gain was reduced in a dose-related manner in males, and in HDF. On Day 29 (just prior to mating), body weight in LD, MD, and HD males were 6, 11, and 17% lower than body weight in CM, and 4% lower in HDF than in CF. Body weight gain was significantly reduced in MD and HD females during the first 2 wks of gestation (Wk 1: 40 and 58%, respectively; Wk 2: 24 and 27%, respectively), and remained lower throughout the rest of gestation (Day 21: 12, 19, and 15% at the LD, MD, and HD, respectively). In general, changes in food consumption were consistent with those noted on body weight.

Mating and fertility data are summarized in the following sponsor's table:

Group	No. Sperm Positive (%)		No. Gravid (%)		No. Non-Gravid
control x control	8/10	(80)	10/10*	(100)	0/10
10 mg M x 10 mg F	10/10	(100)	9/10	(90)	1/10
40 mg M x 40 mg F	8/10	(80)	10/10*	(100)	0/10
160 mg M x 160 mg F	8/10	(80)	5/8	(63)	5/10
control M x 160 mg F	6/9	(67)	2/6	(33)	7/9
160 mg M x control F	10/10	(100)	9/10**	(100)	1/10

* Two females had litters with no positive signs of copulation noted.
** One female died of dystocia.

The pregnancy rate was reduced in the grp in which only the female was treated (at 160 mg/kg). The fertility rate was reduced in this grp and in the grp in which both male and female received the HD. Neither the fertility or pregnancy rate was reduced in the grp in which HDM were mated with CF.

Time-to-mate data were not summarized. According to the sponsor, time-to-mate was prolonged in all grps except for the grp in which treated HDM were mated with CF. The range of mating intervals was as follows: C x C: 1-5 days, LD: 1-14 days, MD: 4-12 days; HDF x CM: 1-10 days, HD: 2-12 days. The number of females with successful mating occurring at Day 10 or later was 2/20, 2/10, 2/10, and 6/20 in CF, LDF, MDF, and HDF, respectively. The sponsor noted that, for HDF who were pregnant (either grp), time to positive sperm smear was 9-12 days. [Although individual estrus data were provided, there was no discussion by the sponsor. It is unknown whether or not all incidences of estrus were recorded (cf. Segment I study)]

Gestation and litter data are summarized in the following sponsor's table:

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TABLE 7

92-72
CP-88,0

MEAN REPRODUCTIVE DATA FOR PREGNANT FEMALES & PUP VIABILITY THROUGH POSTNATAL DAY 10

DOSE		LENGTH OF GESTATION[a]	NO. OF IMPLANTATIONS	MEAN NO. BORN TOTAL	MEAN NO. BORN ALIVE	% BORN DEAD	NO. (%) SURVIVING ON DAY 1	NO. (%) SURVIVING ON DAY 4[b]	NO. (%) SURVIVING ON DAY 7	NO. (%) SURVIVING ON DAY 10
CONTROL	N	8	10	10	10	10	10	10	10	10
	MEAN	21.4	15.8	15.2	14.7	2.9	14.7(100)	14.7(100)	7.9(98.8)	7.9(100)
	S.D.	0.52	2.57	2.5	2.1					
CONTROL X 160.0 MG/KG MALE	N.	8	8	8	8	8	8	8	8	8
	MEAN	21.0	16.5	15.6	15.6	0.0	15.6(100)	15.6(100)	7.9(98.4)	7.9(100)
	S.D.	0.58	1.69	1.9	1.9					
10.0 MG/KG	N.	9	9	9	9	9	9	9[c]	8	8
	MEAN	21.4	14.2	14.0	12.7	8.0	12.7(100)	11.9(84.8)	7.9(98.4)	7.9(100)
	S.D.	0.88	6.45	5.3	5.2					
40.0 MG/KG	N.	8	10	10	10	10	10	10	10[d]	9
	MEAN	21.6	15.1	14.5	11.3	22.4*	11.3(100)	10.1(87.4)	6.7(90.0)	7.4(97.2)
	S.D.	0.92	1.52	1.5	3.6					
160.0 MG/KG	N.	5	5	5	5	5	5	5	5	5
	MEAN	21.6	14.6	13.0	10.0**	24.8**	10.0(100)	8.8(88.2)*	7.4(100)	7.4(100)
	S.D.	0.55	1.14	1.5	2.2					
160.0 MG/KG X CONTROL MALE	N.	2	2	2	2	2	2	2[e]	1	1
	MEAN	21.5	14.0	13.5	7.5	42.9	7.5(100)	8.5(58)*	8.0(100)*	8.0(100)
	S.D.	0.71	0.0	0.7	0.8					

[a]Calculated for those animals with a correctly detected sperm positive vaginal smear.
N = Number of litters.
[b]Calculated for surviving litters only.
[c]No prior to culling
[d]Dam was sacrificed. Entire litter was dead by lactation day 4
[e]Dam was sacrificed. Entire litter was dead by lactation day 5.
[f]Dam was sacrificed. Entire litter was dead by lactation day 3.
* p<0.05, ** p<.01

[There would appear to be an error in the last line of Table 7: on Day 4 postpartum, only 1 litter was left in the HDF x CM grp, i.e., 1 instead of 2.] The length of gestation was similar among grps. However, the number of stillborn pups was increased in all grps except for the C x HDM grp. In addition, a greater no. of those pups born alive were dead by Day 4 postpartum in the same grps. Pup survival was similar among grps (of those alive on Day 4) on Day 7 and 10.

According to the sponsor, pup survival "...appeared to be dependent, in part, on the degree of severity of the sedation in the dams and subsequent lack of maternal care particularly during the time through postnatal Day 4". [Due to insufficient time, individual data were not examined.]

Pup wt was significantly reduced in the HD grp on Days 7 and 10 postpartum (22-28%); however, pup wt tended to be lower in all drug-treated grps except, perhaps, for the CF x HDM grp.

According to the sponsor, no gross pathology findings were detected in either the F_0 or the F_1 generation.

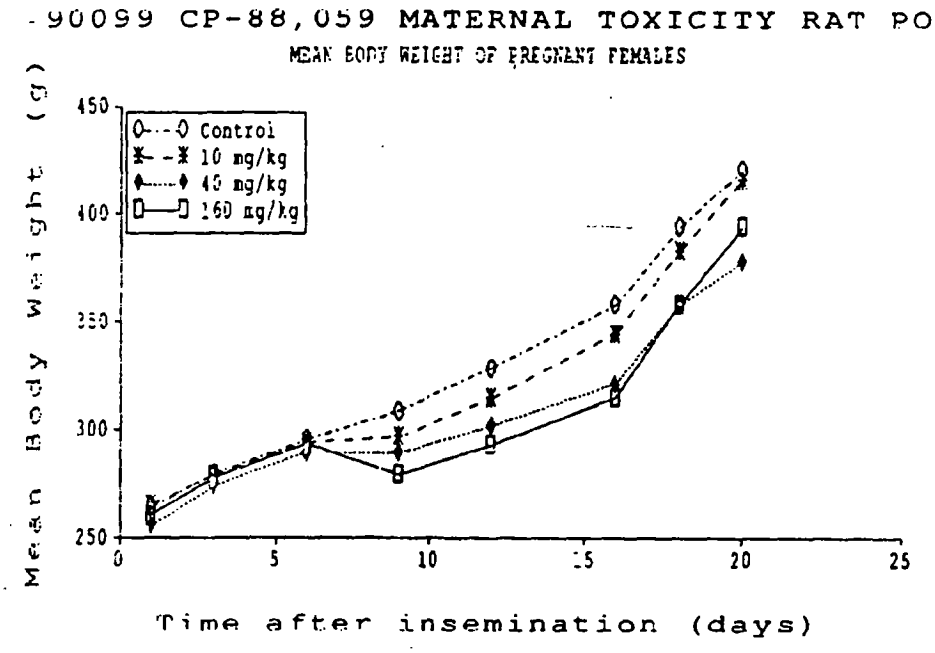
2. **Maternal toxicity study in rats by the oral route** (Protocol no. 90099, Pfizer Central Research, study dates: 11-12/90, report date: 6/6/91, GLP, Vol 1.28)

Methods: CP-88,059 (batch no. 19,561-119-1MS) was administered orally (gavage) to female Sprague-Dawley rats (n = 7/grp) at doses of 0, 10, 40, and 160 mg/kg. CP-88,059 was suspended in 0.5% methylcellulose for dosing. Dosing was once daily from Day 6 to Day 15 post insemination (p.i.). Observations consisted of the following: clinical signs, body wt/food consumption, terminal studies [Day 20 p.i.; examination of ovaries/uteri, no. of fetuses, fetal body wt/sex].

Results: there were no unscheduled deaths. Drug-related clinical signs (summarized in the following sponsor's table) were noted at all doses.

	Control	10 mg/kg	40 mg/kg	160 mg/kg
Ptosis	0/7	0/7	7/7	7/7
Prostration	0/7	7/7	7/7	7/7
Piloerection	0/7	7/7	7/7	7/7
Lacrimation	0/7	1/7	7/7	7/7
Chromodacryorrhoea	0/7	0/7	0/7	2/7
Hunched posture	0/7	0/7	7/7	7/7

Body weight was affected at all doses, although minimally at the LD. By Day 9 of gestation (i.e., p.i.), body weights in LD, MD, and HD females were 4, 6, and 10% lower than in CF. At the end of the dosing period, body wts were 4, 10, and 12%, respectively, lower than CF. At sacrifice, body wt was still reduced in MD and HD females. Changes in body wt are illustrated in the following sponsor's figure (**note that ordinate does not start at zero**):



The body weight gain data were summarized in the following sponsor's table:

	Control	10 mg/kg	40 mg/kg	160 mg/kg
Days 6-9 p.i.	13.1	2.4	-0.5	-14.2
Days 9-12 p.i.	19.7	17.3	11.9	13.7
Days 12 16 p.i.	29.9	30.6	19.8	21.9
Days 16 21 p.i.	62.0	71.3	57.1	79.2

Food consumption was reduced on Day 9 and 16 of gestation in MDF and HDF (12-21 and 23-26%, respectively); at the LD, food consumption was reduced (8%) only on Day 9.

The litter data are summarized in the sponsor's table below. There were no significant or consistent (i.e., dose-related) effects on any of the parameters assessed.

RESULTS

	CONTROL	10 MG/KG	40 MG/KG	160MG/KG
REPRODUCTIVE VARIABLES FOR SACRIFICED FEMALES				
PREGNANCY RATE (%)	7/ 7 (100)	7/ 7 (100)	7/ 7 (100)	7/ 7 (100)
VIABLE LITTERS ON DAY 20 (%)	7/ 7 (100)	7/ 7 (100)	7/ 7 (100)	7/ 7 (100)
CORPORA LUTEA MEAN+S.D.	18.9± 2.48	18.1± 1.21	17.1± 1.68	18.4± 2.99
IMPLANTATION SITES MEAN+S.D.	15.6± 2.23	16.4± 1.27	16.3± 1.11	17.1± 2.54
NO FOETUSES MEAN+S.D.	14.4± 1.72	16.0± 2.00	15.4± 0.98	16.3± 2.50
IMPLANTATION RATE (%)	109/132(82.6)	115/127(90.6)	114/120(95.0)	120/129(93.0)
EMBRYONORTALITY RATE (%)	8/109(7.3)	3/115(2.6)	6/114(5.3)	6/120(5.0)
FOETAL DEVELOPMENT				
SEX RATIO M/F(%)	50/ 51(98)	57/ 55(104)	46/ 62(74)	61/ 53(115)
MEAN FOET.WEIGHTS MALES (g)	3.69± 0.27	3.72± 0.31	3.60± 0.48	3.61± 0.23
MEAN FOET.WEIGHTS FEMALES (g)	3.52± 0.32	3.55± 0.23	3.31± 0.46	3.45± 0.19
MEAN BODY WEIGHT GAIN OF PREGNANT FEMALES (grams)				
NO OF PREGNANT FEMALES	7	7	7	7
FROM DAY 1 TO DAY 20	156.3	150.4	122.9	133.4
FROM DAY 1 TO DAY 6	31.5	28.8	34.5	32.8
FROM DAY 6 TO DAY 16	62.7	50.3	31.3	21.4
FROM DAY 16 TO DAY 20	62.0	71.3	57.1	79.2

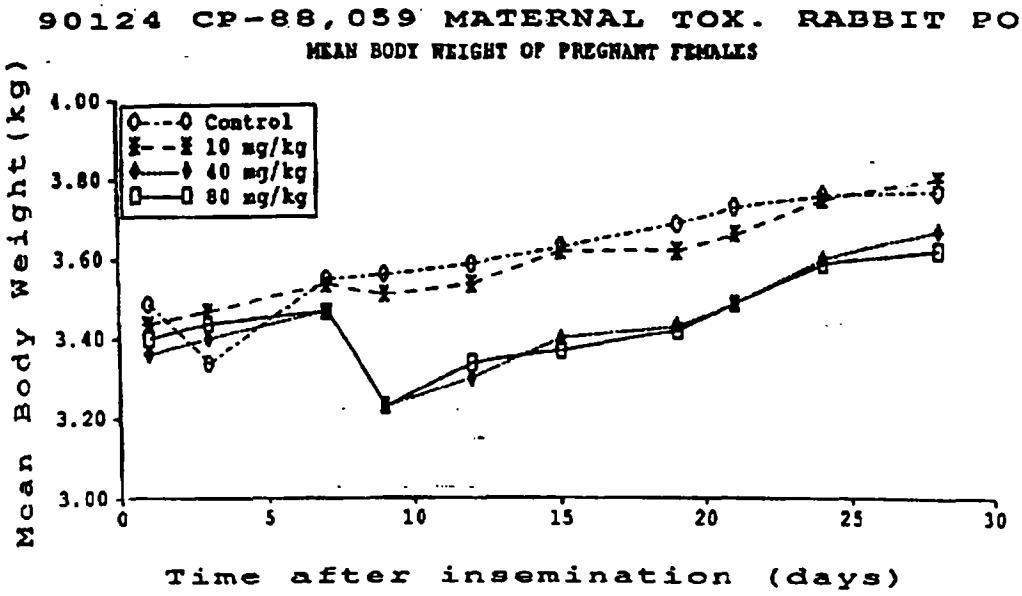
3. **Maternal toxicity study in rabbits by the oral route** (Protocol no. 90124, Pfizer Central Research, report date: 6/91, study dates: 11-12/90, GLP, Vol 1.28)

Methods: CP-99,059 (batch no. 19,561-119-1MS) was administered orally (gavage) to New Zealand White rabbits (n = 7/grp) at doses of 0, 10, 40, and 80 mg/kg from Day 7 to Day 18 of gestation. CP-88,059 was suspended in 0.5% methylcellulose for dosing. Observations consisted of the following: clinical signs, body wt/food consumption, terminal studies [Day 28 of gestation; uteri/ovaries examination, litter data (implantation rate, no. of corpora lutea), no. of fetuses, fetal wt/sex].

Results: there were no unscheduled deaths. According to the sponsor, coryza was observed in all grps, including CF (4-5/grp). Abnormal head position ("...bowed to the right") was noted in 1 HDF; absence of feces and/or urine was noted in 2/7 HDF. The pregnancy rate (i.e., the

no. of females with at least 1 live fetus) was low in all grps (3-4/grp), including CF. One female in each of the MD and HD grps aborted the entire litter; both occurred on Day 19 of gestation.

Body wt was affected at the MD and HD; these data are illustrated in the sponsor's figure below (**note that the ordinate does not start at zero**). The maximum reduction in body wt (9% relative to CF) occurred on Day 9 of gestation. By Day 28, body weights were only 3-4% lower in these grps as compared to CF.



The food consumption data were not summarized. According to the sponsor, reduced and/or absent food intake occurred in all grps; however, all MDF and HDF were affected (3/7 CF, 2/7 LD).

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

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RESULTS

	CONTROL	10 MG/KG	40 MG/KG	80 MG/KG
REPRODUCTIVE VARIABLES FOR SACRIFICED FEMALES				
PREGNANCY RATE (%)	5/ 7 (71)	4/ 7 (57)	4/ 7 (57)	5/ 7 (71)
VIABLE LITTERS ON DAY 28 (%)	4/ 5 (80)	4/ 4 (100)	3/ 4 (75)	4/ 5 (80)
CORPORA LUTEA MEAN±S.D.	7.8± 1.71	8.3± 0.96	6.7± 1.15	9.8± 1.71
IMPLANTATION SITES MEAN±S.D.	7.5± 2.08	5.8± 2.63	6.7± 1.15	8.3± 2.87
NO FCETUSES MEAN±S.D.	7.5± 2.08	5.8± 2.63	6.0± 1.00	7.0± 2.45
IMPLANTATION RATE (%)	30/ 31(96.8)	23/ 33(69.7)	20/ 20(100.0)	33/ 39(84.6)
EMBRYOMORTALITY RATE (%)	0/ 30(0.0)	0/ 23(0.0)	2/ 20(10.0)	5/ 33(15.2)
FOETAL DEVELOPMENT				
MEAN FCET.WEIGHTS	28.19± 3.40	32.79± 5.99	33.63± 3.73	27.44± 4.59
MEAN BODY WEIGHT GAINS OR LOSSES OF PREGNANT FEMALES (grams)				
NO OF PREGNANT FEMALES	4	4	3	4
FROM DAY 1 TO DAY 28	277.3	358.5	310.3	227.3
FROM DAY 1 TO DAY 7	56.0	100.3	113.7	77.0
FROM DAY 7 TO DAY 19	146.8	77.0	-36.7	-52.3
FROM DAY 19 TO DAY 28	74.5	181.3	233.3	202.5

The only dose-related finding was an increase in the embryomortality (i.e., no. of dead implants/no. of implantation sites) at the MD and HD (10 and 15.2% vs 0% for CF and LDF). The no. of fetus/litter and fetal body wt were similar among grps.

There were no apparent drug-related findings in dams at necropsy.

4. **Reproductive study III, teratology by the oral route in New Zealand White rabbits** (Study no. 94-720-30, Pfizer Central, report date: 2/96, study dates: 11/9/94-12/15/94, GLP, Vol 1.28)

Animals: New Zealand White rabbits
initial age: not specified
initial body wt: 3.29 kg on Day 6 of gestation
n = 24 CF, 20 LDF, and 24 HDF; the no. of satellite TK animals was not specified. From the individual data tables, it would appear to be 4 for C and HD grps.

Drug: CP-88,059 (lot no. 31081-79-1F)
vehicle: 0.5% methylcellulose
stability: stability and homogeneity data were not provided. According to the sponsor, CP-88,059 was stable at rm temperature for 7 days in vehicle. Also according to the sponsor, achieved drug concentrations were within 15% of intended, except for the 5.0 mg/mL suspension (used for the LD) which was only 82% of intended.
doses: 0, 10, 30 mg/kg
route: p.o. (gavage)
dosing volume: 2 mL/kg
duration: Day 6 through Day 18 of gestation; TK animals were dosed through Day 19.

Observations

Clinical signs: all rabbits were observed twice daily.

Body weight: body wts were recorded daily.

Food consumption: food consumption was recorded daily.

Terminal studies

Gross pathology: all dams were sacrificed on Day 28 of gestation. A complete necropsy was not performed. Uteri and ovaries were examined. The following parameters were recorded: no. of corpora lutea, uterine wt, implantation sites.

TK: Blood samples were collected at 1, 3, and 5 hr following a 30 mg/kg dose of [^{3}H]CP-88,059. Animals were sacrificed after the last blood collection and 3-4 fetuses/dam were collected for analysis of fetal drug levels. Plasma and tissue radioactivity were quantitated using . The no. of animals for TK analysis was not specified in the Methods section.

Fetal examination: all fetuses were examined for external malformations. Visceral examinations were performed using the Stuckhardt and Poppe methodology. Skeletal examinations were conducted using alcian blue and alizarin red stains. Fetuses with variations or malformations in bone used to assess degree of ossification were not included in the analysis of ossification index or rib pair counts. Ossification was noted as follows: N = normal, S = slight delay, M = marked delay, P = ossification present, A = ossification absent.

Results

Mortality: there were 2 unscheduled deaths, both at the HD. One death resulted from a dosing accident (Day 13) and 1 HDF (#60) was sacrificed moribund on Day 22 of gestation.

Clinical signs: summary or individual data were not provided. According to the sponsor, drug-related clinical signs were observed only at the HD; these consisted of decreased activity (5/24 HDF) and "loose matted stool" (2/24 HDF).

Body weight: there were no drug-related findings.

Food consumption: there were no drug-related findings.

TK: the C_{max} values for total plasma radioactivity ranged from 5 µg-eq/mL. Fetal tissue levels ranged from µg-eq/gm (these are 5-hr values). The ratio of maternal plasma to fetal tissue levels at 5 hr postdosing was ≈0.3.

Terminal studies

Litter data: the data are summarized in the following sponsor's Table 4:

TABLE 4
MEAN REPRODUCTIVE PARAMETERS

DOSE		CORPORA LUTEA	IMPLANTATION SITES	VIABLE FETUSES	RESORPTIONS EARLY	RESORPTIONS LATE	DEAD FETUSES	MEAN FETAL WEIGHT (G)		MEAN PLACENTAL WEIGHT (G)		SEX RATIO
								Male	Female	Male	Female	
Control	No.	19	19	19	19	19	19	19	19	19	19	
	Mean	9.6	9.4	8.6	0.3	0.5	0.0	35.85	33.93	5.16	4.88	1.43
	s.d.	1.7	1.8	1.7	0.7	1.4	0.0	3.81	3.59	0.76	0.71	
10.0 mg/kg	No.	20	20	20	20	20	20	20	18	20	18	
	Mean	8.0	8.0	7.7	0.2	0.1	0.0	36.35	36.2[illegible]	5.25	4.91	1.20
	s.d.	1.9	1.9	1.9	0.4	0.3	0.0	4.18	5.02	0.82	0.78	
30.0 mg/kg	No.	19	19	18	18	18	18	18	18	18	18	
	Mean	8.8	8.7	8.0	0.1	0.2	0.0	36.86	34.95	5.36	5.00	1.16
	s.d.	1.7	1.8	1.6	0.2	0.5	0.0	4.44	4.77	0.95	0.87	

There were no apparent drug-related effects on any of the parameters evaluated.

Fetal examinations: a total no. of 19, 20, and 18 litters were examined from CF, LDF, and HDF, respectively. [Of the original n/grp, 4 CF and 4 HDF were used for TK analysis, 1 CF was not pregnant, and 2 HDF died or were sacrificed moribund.]

External findings consisted of vestigial tail with spina bifida in 2 C fetuses from 1 litter (#9) and "runt" in 1 C, 1 LD, and 2 HD litters.

The visceral findings are summarized in the sponsor's table below. The incidence of renal pelvis dilation was increased at the HD, both in terms of affected fetuses and litters. In addition, ventricular septal defect was noted in 3 HD litters (1 affected fetus/litter). One of these fetuses exhibited a panoply of findings: diaphragmatic hernia, stomach and liver lobe in thoracic cavity, dextrocardia, left atrium reduced ventricular septal defect, left atrial septal defect, left portion of bicuspid papillary muscle absent, left lobe of lung absent, right lung lobes reduced, liver lobes misshaped, ectopic left kidney, and left kidney and ureter moderately dilated. [Historical data supplied by the sponsor is attached.]

TABLE 6
VISCERAL EXAMINATION OF FETUSES

84-720-30
CP-88.059-1

	CONTROL F	CONTROL L	10.0 MG/KG F	10.0 MG/KG L	30.0 MG/KG F	30.0 MG/KG L
TOTAL NO. EXAMINED	163	19	153	20	144	18
THORAX						
ACCESSORY LUNG LOBE ABSENT	3	3	5	4	1	1
GLOBULAR SHAPED HEART, LEFT VENTRICULAR WALL THICKENED, VENTRICULAR SEPTAL DEFECT	0	0	0	0	1	1
RIGHT VENTRICLE AND PULMONARY ARTERY ENLARGED, VENTRICULAR SEPTAL DEFECT	0	0	0	0	1	1
DIAPHRAGMATIC HERNIA DEXTROCARDIA VENTRICULAR SEPTAL DEFECT ATRIAL SEPTAL DEFECT PORTION OF BICUSPID PAPILLARY MUSCLE ABSENT, LEFT LOBE OF LUNG ABSENT RIGHT LUNG LOBES REDUCED ECTOPIC LEFT KIDNEY	0	0	0	0	1	1
ABDOMEN						
RENAL PELVIS DILATED	0	0	0	0	6	3
RIGHT TESTIS ABSENT	0	0	1	1	0	0

APPEARS THIS WAY
ON ORIGINAL

CUMULATIVE LABORATORY DATA IN NEW ZEALAND WHITE RABBIT
1991 TO 1994

SUPPLIER: [redacted]

Year		1991	1992			1993		1994
Studies		1	2	3	4	5	6	7
No. Fetuses Examined		84	174	309	78	854	812	830
No. Litters Examined (L)		12	22	37	8	104	71	70
Globular Shaped Heart	No. Fetuses W/ Finding (L)	1 (1)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (1)
	%	0.93	0.00	0.00	0.00	0.00	0.00	0.15
	Range							
Dextrocardia	No. Fetuses W/ Finding (L)	0 (0)	1 (1)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
	%	0.00	0.50	0.00	0.00	0.00	0.00	0.00
	Range							
Diaphragmatic Hernia	No. Fetuses W/ Finding (L)	0 (0)	1 (1)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
	%	0.00	0.50	0.00	0.00	0.00	0.00	0.00
	Range							
Ventricular Septal Defect	No. Fetuses W/ Finding (L)	0 (0)	0 (0)	0 (0)	0 (0)	3 (3)	0 (0)	1 (1)
	%	0.00	0.00	0.00	0.00	0.34	0.00	0.15
	Range							
Enlarged Atrium	No. Fetuses W/ Finding (L)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (1)	0 (0)
	%	0.00	0.00	0.00	0.00	0.00	0.00	0 (0)
	Range							
Enlarged Ventricle	No. Fetuses W/ Finding (L)	0 (0)	0 (0)	0 (0)	0 (0)	2 (1)	0 (0)	0 (0)
	%	0.00	0.00	0.00	0.00	0.21	0.00	0 (0)
	Range							
Malformation of Heart and Vessels	No. Fetuses W/ Finding (L)	0 (0)	0 (0)	0 (0)	0 (0)	1 (1)	0 (0)	0 (0)
	%	0.00	0.00	0.00	0.00	0.12	0.00	0.00
	Range							

Some of the results of the skeletal examinations were expressed as an "index" and not as incidences (fetal or litter) and, therefore, could not be compared to the individual data. The sponsor has been asked to provide summaries expressed as incidences of affected fetuses and litters. According to the sponsor's summary table [data expressed in terms of an "ossification index" (mean ± SD) or number of affected fetuses], there were no drug-related effects.

5. **Teratology (Study III) by the oral route in New Zealand White rabbits** (Study no. 95-720-34, Pfizer Central Research, report date: 2/96, study dates: 5/8/95-6/8/95, GLP, Vol 1.29)

Animals: New Zealand White rabbit[ ]
mean body weight: 3.78 kg on Day 6 of gestation
diet/water: rabbit chow (150 gm/day); purified tap water. Apparently, no analysis of food or water was conducted. The sponsor stated that "To the best of our knowledge, there were no contaminants in the diet or water that could be expected to interfere with the outcome of the study."
n = 30/grp

Drug: CP-88,059 (lot no. 31081-79-1F)
vehicle: 0.5% methylcellulose
stability/homogeneity: the sponsor stated that CP-88,059 was stable in suspension for 7 days; no supportive data were provided. Achieved concentrations were said to be within 15% of intended; again, however, no supportive data were provided.
doses: 0 and 30 mg/kg
duration: Day 6 through Day 18 of gestation
dosing volume: 2 mL/kg

Observations

Clinical signs: animals were observed twice daily.

Body wt: body wts were recorded daily.

Food consumption: food intake was recorded daily.

Terminal studies

Gross pathology: animals were sacrificed on Day 28 of gestation. A complete necropsy was not performed. Uteri and ovaries were examined for corpora lutea and implantation sites. Uteri and placenta were weighed. The no. of fetuses (live/dead) were recorded.

Fetal examination: all fetuses were examined for external and visceral (using Stuckharde and Poppe, 1984) findings.

Results

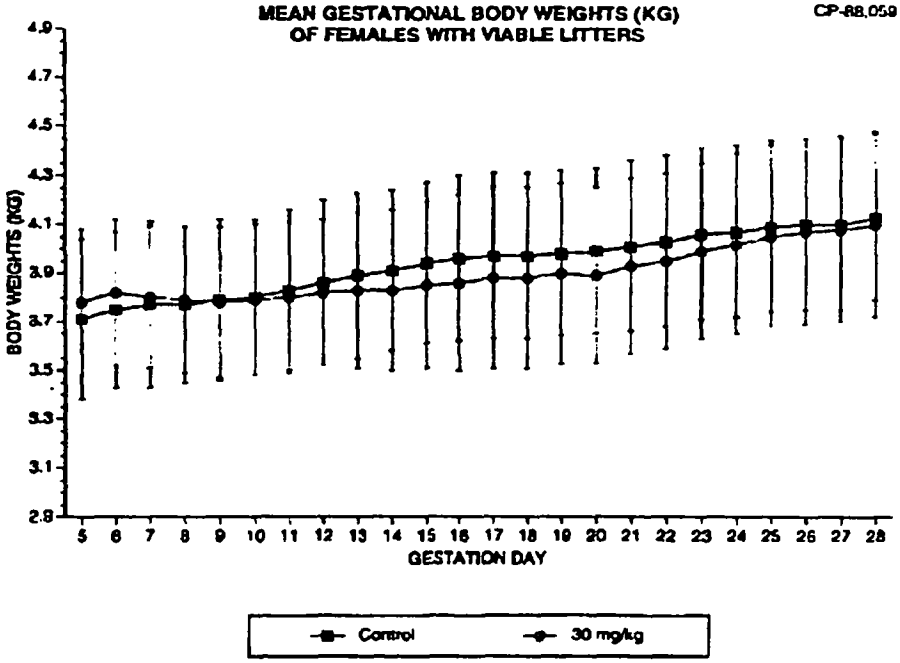
Mortality: there was one unscheduled sacrifice (1 DTF); this animal was sacrificed moribund on Day 27.

Clinical signs: The only drug-related clinical sign was loose or soft stools; this finding was observed in 6/30 DTF.

Body weight: body weight changes are illustrated in the sponsor's Figure 1 provided below.

FIGURE 1

95-720-34
CP-88,059



There were no significant differences in body wt between grps. Body wt gain, however, was affected in DTF. During Days 6-12 of gestation, there was a mean body wt loss of 0.01 ± 0.08 kg versus a mean gain of 0.11 ± 0.07 kg in CF.

Food consumption: food consumption was reduced in DFT by 7 and 20% during Days 6-12 and 13-19 of gestation, respectively. During the last wk of gestation, food consumption was similar between grps.

Terminal studies

Gross pathology: in the DTF sacrificed moribund (#53), "...pale liver, ...no food in stomach..." and dehydration were detected. The only other findings noted only in DTF were fluid filled cysts "associated with" an oviduct or fallopian tube and red fluid in right uterine horn.

Litter parameters: the data were summarized in the sponsor's Table 4 given below. There was no significant drug-related effects on any of the parameters assessed. Fetal body wt did, however, tend to be lower in DTF (≈4%) as did placental wt (9-10%).

TABLE 4

95-720-34
CP-88,059

MEAN REPRODUCTIVE PARAMETERS

Dose		Corpora Lutea	Implantation Sites	Viable Fetuses	Resorptions Early	Resorptions Late	Dead Fetuses
Control	No.	28	28	28	28	26	28
	Mean	11	10.04	9.18	0.39	0.39	0.07
	s.d.	1.5	1.6	2.2	1.2	0.7	0.3
30 mg/kg	No.	26	26	26	26	26	26
	Mean	11	10.31	9.85	0.12	0.50	0.04
	s.d.	2.0	2.20	2.06	0.33	1.03	0.20

MEAN FETAL AND PLACENTAL WEIGHTS

Dose		Mean Fetal Weight (G)		Mean Placental Weight (G)		Sex Ratio
		Male	Female	Male	Female	
Control	No.	28	28	28	26	
	Mean	35.84	35.14	5.40	5.16	0.97
	s.d.	3.85	3.59	0.77	0.73	
30 mg/kg	No.	26	26	26	26	
	Mean	35.15	34.85	4.85	4.69	1.25
	s.d.	3.91	4.40	0.61	0.83	

Fetal examinations: visceral findings were summarized in the Sponsor's Table 5 provided below. It should be noted that the sponsor did not provide a summary table of malformations.

VISCERAL EXAMINATION OF FETUSES

	Control		30 mg/kg	
	F	L	F	L
TOTAL NO. EXAMINED	256	28	252	26
TOTAL NORMAL	247	19	241	16
HEAD				
RED AREA AROUND EYE	0	0	1	1
DILATED THIRD VENTRICLE	0	0	1	1
THORAX				
ACCESSORY LOBE OF LUNG ABSENT	5	5	6	5
CLEFT IN APEX OF HEART	1	1	1	1
VENTRICULAR SEPTAL DEFECT	1	1	1	1
GLOBULAR SHAPED HEART, ENLARGED AORTA	0	0	1	1
ABDOMEN				
FLUID FILLED SAC ASSOCIATED WITH OVARY	2	2	1	1
FLUID FILLED SAC ASSOCIATED WITH TESTES	1	1	0	0
GALL BLADDER ABSENT	0	0	1	1
ECTOPIC KIDNEY	0	0	3	3
KIDNEY REDDENED	0	0	1	1
SMALL OVARY	0	0	1	1
ECTOPIC TESTIS	0	0	1	1

(F)etuses and (L)itters affected per group

Unfortunately, the fetuses from DTF #53 (sacrificed moribund on Day 27 of gestation) were apparently not examined. Also, dilated third ventricle was detected in 1 DT fetus (doe #54). On external examination, exencephaly was detected in 1 DT fetus (doe #39) and "left bent paw" was detected in 1 DT fetus (doe #45).

6. **Reproductive study II, prenatal and postnatal development, in Sprague-Dawley rats** (Study no. 93-720-28, Pfizer Central Research, report date: 3/95, study dates: 9/26/93-1/21/94, GLP, Vol 1.29)

Animals: Sprague-Dawley rats
initial body wts: not specified
initial age: not specified
diet/water: *ad lib*; the sponsor did not provide diet/water analyses, but did state that "To the best of our knowledge, there were no contaminants in the diet or water that could be expected to interfere with the outcome of the study."
n = 20/grp

Drug: CP-88,059-1 (lot no. 23,638-951-1F)
vehicle: 0.5% methylcellulose
storage: drug suspension was stored at rm temperature, protected from the light.
stability: no data were submitted to document stability. According to the sponsor, CP-88,059-1 is stable in suspension for 7 days at rm temperature.
drug formulation: no data were submitted to document drug concentration/homogeneity. According to the sponsor, drug suspensions were analyzed during Wks 1, 2, 4, 7, and 10, and that all were within 15% of

intended concentration and "...were considered homogenous." Suspensions were prepared weekly.
doses: 0, 5, 10, and 40 mg/kg
route: p.o. (gavage)
duration: Day 6 of gestation through Day 21 of lactation

Observations

F_0 generation

Clinical signs: all dams were observed twice daily.

Body weights: body wts were recorded daily from Day 6 of gestation through Day 21 of lactation.

Food consumption: food intake was recorded daily throughout gestation.

Reproductive parameters: all dams were allowed to deliver naturally. Day of parturition was designated Day 0. The no. of live/dead pups were recorded for each litter.

Terminal studies

Gross pathology: dams were sacrificed on Day 21 postpartum and necropsied. Dams that "...had litters that did not meet criteria..." were killed and were not included in the study. These "criteria" were as follows: (1) ≥2 pups/sex/litter and (2) tot no. of pups in litter had to be ≥7.

Dams that were not pregnant were sacrificed on Day 24 of gestation, and reproductive organs were examined.

F_1 generation (not dosed directly)

Body wts: pup body wt was recorded on Days 4, 7, 10, 14, and 21 postpartum.

Culling: on Day 4 postpartum, litters were culled to 4/sex/litter. If a litter had <4 of either males or females, additional pups of the available sex were retained in order to have a total of 8 pups/litter.

On Day 21 postpartum, 2/sex/litter were selected for assessment of developmental landmarks.

Postnatal development: the following tests were performed on all pups: surface righting (starting Day 1 postpartum), incisor eruption (starting Day 7 postpartum), eye opening (starting Day 10 postpartum), air righting (starting Day 14 postpartum), visual cliff avoidance (starting Day 14 postpartum). For all but the latter test, pups were tested until a positive response was obtained. For the visual cliff avoidance test, testing was continued on Day 21 in the selected 2/sex/litter.

The following tests were conducted in the 2/sex/litter selected on Day 21: vaginal opening (starting on Day 28 postpartum), preputial separation (starting on Day 35 postpartum), auditory function (Day 30 postpartum), FOB (Day 21), visual system integrity by ophthalmoscope (once per pup, Day 21-28), motor activity (Day 23 postpartum), Cincinnati Water Maze (Days 55-65 postpartum),

passive avoidance (Days 63-70 postpartum).

Terminal studies

Gross pathology: pups culled on Day 4 postpartum were sacrificed and examined for external and internal abnormalities.

Pups not selected for additional study on Day 21 postpartum were sacrificed and necropsied.

Pups selected for learning and memory assessment were sacrificed at the end of the tests (≈Day 70 postpartum) and necropsied.

Statistics

In most analyses, the litter was considered the experimental unit.

Results

F_0 generation

Mortality: there were no unscheduled deaths.

Clinical signs: no summary table was provided. According to the sponsor, sedation and chromodacryorrhea were observed at all doses, with incidence and/or severity being dose-related.

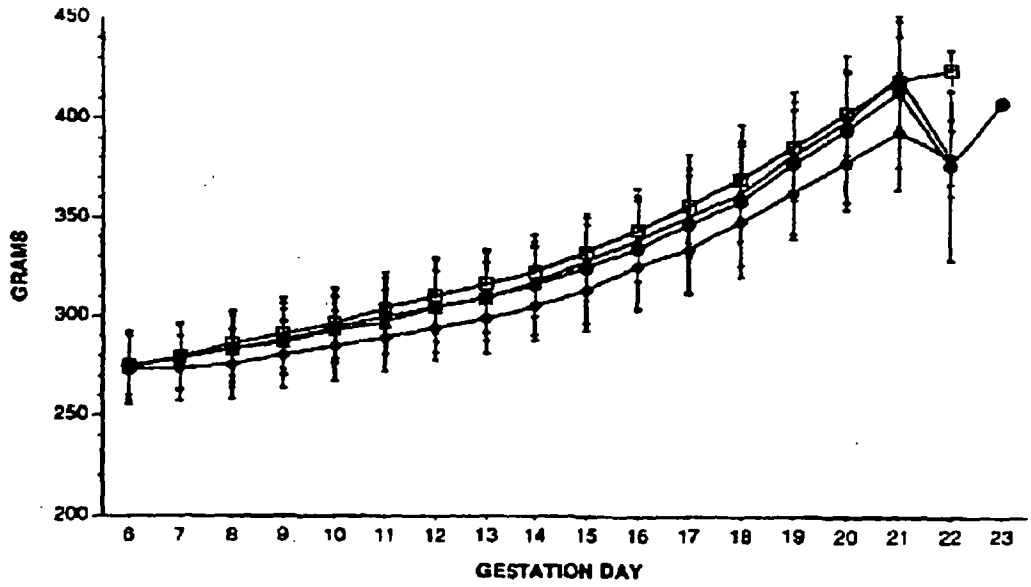
Body weight: body weight changes during gestation and lactation are illustrated in the sponsor's figures below. [Note that the ordinate does not start at zero.] During gestation, body wt was significantly lower in HDF (as compared to CF) from Days 10 through 21 (4-6%). On Day 22 of gestation, there were no significant differences among grps; however, mean body wt was ≈10% lower in all treated grps as compared to CF. Body wt gain data during gestation are summarized in the following sponsor's table:

Body Weight Gains (g) of Dams During Gestation

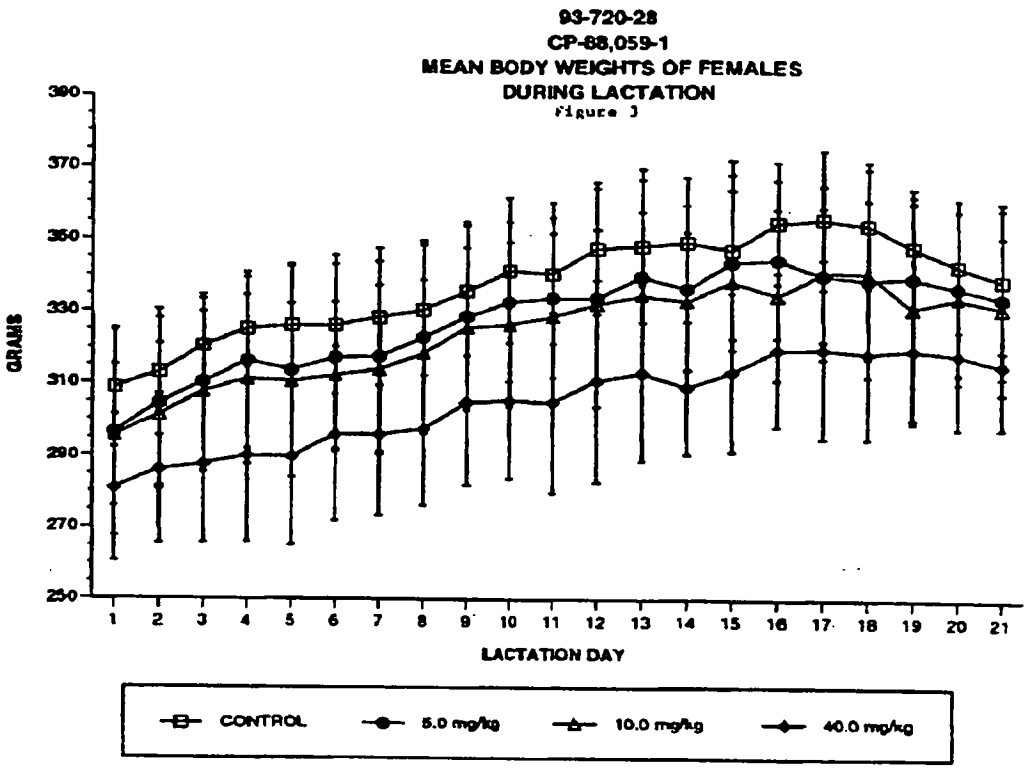
Gestation Days	Control	5 mg	10 mg	40 mg
6-12	35.7±10.3	32.0±15.0	30.5±8.1	21.3±9.9**
12-20	108.8±16.4	109.2±22.7	117.2±14.1	99.8±22.0
6-20	144.6±17.1	141.2±30.8	147.7±20.3	121.1±17.6**

** $p < .01$

93-720-28
CP-88,059-1
MEAN GESTATIONAL BODY WEIGHTS OF
FEMALES WITH VIABLE LITTERS
Figure 1



During lactation, body wt tended to be lower in treated grps (as compared to CF), with the effect being significant primarily in HDF (≤12%). Body wt gain was somewhat lower in treated grps during Days 8-14 of lactation and in HDF during Days 1-14 (24-35%); however, by the last wk of lactation body wt gain was noted only in HDF.



Body wt gain data during lactation are summarized in the following sponsor's table:

Body Weight Gains (g) of Dams During Lactation

Lactation Days	Control	5 mg	10 mg	40 mg
1-7	19.9±14.2	21.2±16.1	18.4±12.6	15.2±12.6
8-14	19.1±10.4	13.8±12.9	14.7±10.7	12.5±10.5
15-21	-8.3±20.4	-9.8±14.2	-7.0±13.0	2.0±9.1*

* $p < .05$

Food consumption: food intake was similar among grps during gestation.

Reproductive parameters: the sponsor's summary table is given below. According to this summary, there were no drug-related effects on length of gestation, implantation sites, or the total number of pups/litter. However, the no. of stillborn pups was increased and the pup survival rate at Day 4 was reduced in HDF. The no. of stillborn pups was also increased in LDF and MDF (the effect was dose-related); however, the effect at these doses were not statistically significant.

MEAN REPRODUCTIVE DATA FOR PREGNANT FEMALES AND PUP VIABILITY THROUGH POSTNATAL DAY 21

		LENGTH OF GESTATION	IMPLANTATION SITES	TOTAL BORN	NO. PUPS AT BIRTH DEAD	BORN ALIVE	ALIVE DAY 4	ALIVE DAY 21
CONTROL	N	20	20	20	20	20	20	20
	MEAN	21.2	15.8	14.5	0.0	14.5	14.4	8.0
	S.D.	0.4	1.8	1.4	0.0	1.4	1.5	0.0
5.0 MG/KG	N	20	20	20	20	20	20	20
	MEAN	21.3	15.9	14.2	0.2	14.0	13.8	7.9
	S.D.	0.6	1.8	2.3	0.7	2.3	2.6	0.5
10.0 MG/KG	N	18	18	18	18	18	18	18
	MEAN	21.1	15.7	14.5	0.4	14.1	13.7	8.0
	S.D.	0.3	2.1	1.8	0.8	1.9	2.0	0.0
40.0 MG/KG	N	20	20	20	20	20	20	20
	MEAN	21.3	14.7	14.0	1.3**	12.8	11.6**	8.0
	S.D.	0.5	1.5	1.8	1.0	2.2	2.3	0.0

**p < .01

However, the sponsor's data table did not include data for all pregnant dams. As noted in the Observations section (taken from the sponsor's Methods section), dams not meeting the "criteria" were excluded from the analysis. The following table has been generated using data from all pregnant dams; the data are given as litter means (ranges):

DOSES (mg/kg)	N	LENGTH OF GESTATION (days)	TOTAL BORN	STILLBORN	BORN ALIVE
0	20	21.2	14.4	0	14.4
5	20	21.2	14.2	0.2 (0-3)*	14.0
10	22	21.3	13.1	1.2 (0-8)	12.0
40	21	21.3	13.9	1.3 (0-6)	12.6

*3 deaths in one litter

The only substantive difference when the data from all pregnant dams are included in the summary is an increase in stillborn pups in MDF. This is due to the fact that the highest number of stillbirths were in the MDF that did not meet litter criteria ("NMC"). The sponsor's individual data for the MD grp are presented below:

10.0 MG/KG

INDIVIDUAL REPRODUCTIVE DATA OF FEMALES
AND PUP VIABILITY THROUGH POSTNATAL DAY 21

DAM NO.	GRAVID	LENGTH OF GESTATION	IMPLANTATION SITES	NO. PUPS AT BIRTH			ALIVE DAY 4	ALIVE DAY 21
				TOTAL	DEAD	ALIVE		
47		22	11					
48		22	15					
49		21	19					
50		21	14					
51	NMC	23						
52		21	16					
53		21	14					
54		21	18					
55	NMC	22						
56		21	14					
57	NMC	22						
58		21	13					
59		21	16					
60		21	17					
61		21	16					
62		21	17					
63		21	17					
64		21	13					
65		21	17					
66	NMC	22						
67		21	19					
68		21	16					
69	NG	24						

Dams with litters that were not needed for the study did not have any data reported out

NG = non-gravid
NMC = litter criteria not met

Terminal studies

Gross pathology: no summary or individual tables were provided. According to the sponsor, there were no gross pathology findings in dams.

F_1 generation

Pup survival: the data were summarized in the sponsor's Table 4 (provided above). It should be noted that dams not meeting "litter criteria" (i.e., 4 MDF, 1 HDF) were not included in the survival analysis, except that the Day 4 (but not Day 21) survival data included the data for the 1 HDF.

Clinical signs: there were no apparent drug-related findings.

Body weight: birth wts were not provided. On Day 1, body wt was reduced in MD and HD animals (M-F: 4-5 and 10-12%, respectively) compared to Cs. Summary data are provided in the attached sponsor's Tables 5-6. Body weight was reduced (compared to C pups) throughout the rest of the lactation period in HD pups (9-16% in HDM pups and 12-17% in HDF pups). At the MD, body wt was reduced on Days 14 and 21 in males and from Day 7 on in females. At the LD, body weight was slightly (6%) lower (than C pups) only on Day 14.

After weaning (>Day 21), body weight continued to be reduced in HD pups up through Day 50 in males (10-5%) and Day 36 in females (9-6%).

Table 5

93-720-28
CP-88,059-1

MEAN BODY WEIGHTS (G) OF F1 PUPS: DAYS 1-21

DOSE		DAY 1	DAY 4a	DAY 4b	DAY 7	DAY 10	DAY 14	DAY 21c
MALE								
	N	20	20	20	20	20	20	20
Control	MEAN	6.93	10.31	10.29	16.98	25.47	37.68	59.81
	S.D.	0.40	0.65	0.69	1.28	1.99	2.65	5.93
	N.	20	20	20	20	20.00	20	20
5.0 mg/kg	MEAN	6.80	10.12	10.08	16.79	24.88	35.57*	58.91
	S.D.	0.61	0.99	1.00	1.60	2.18	2.93	4.94
	N.	18	18	18	18	18	18	18
10.0 mg/kg	MEAN	6.62*	10.01	10.01	16.30	24.18	34.76**	57.32
	S.D.	0.43	0.73	0.68	0.97	1.71	2.45	4.81
	N.	20	20	20	20	20	20	20
40.0 mg/kg	MEAN	6.25*	9.25**	9.34**	15.35**	21.89**	31.63**	51.97**
	S.D.	0.39	1.03	1.05	1.61	2.42	3.37	5.58
FEMALE								
Control	N.	20	20	20	20	20	20	20
	MEAN	6.52	9.87	9.89	16.21	24.17	36.51	58.80
	S.D.	0.31	0.68	0.60	1.18	2.14	2.64	5.43
5.0 mg/kg	N.	20	20	20	20	20	20	20
	MEAN	6.41	9.75	9.78	16.18	23.90	34.38*	57.07
	S.D.	0.60	0.96	0.99	1.77	2.29	3.11	4.77
10.0 mg/kg	N.	18	18	18	18	18	18	18
	MEAN	6.22*	9.42	9.41	15.10*	22.65*	32.05**	54.25**
	S.D.	0.46	0.99	1.05	1.18	2.03	2.58	5.05
40.0 mg/kg	N.	20	20	20	20	20	20	20
	MEAN	5.76**	8.72**	8.70**	14.29**	20.58**	30.22**	49.98**
	S.D.	0.46	0.89	0.91	1.49	2.34	3.36	5.59

Table 6

93-720-28
CP-88,059-1

MEAN BODY WEIGHTS (G) OF F1 PUPS : DAYS 29-64

		DAY 29	DAY 36	DAY 43	DAY 50	DAY 57	DAY 64
MALE							
	N	20	20	20	20	20	20
CONTROL	MEAN	112.1	179.0	252.0	322.2	384.9	427.1
	S.D.	9.8	13.2	18.5	24.2	30.1	34.3
	N	20	20	20	20	20	20
5.0 mg/kg	MEAN	110.9	176.8	249.6	318.6	380.1	422.1
	S.D.	8.1	13.1	17.0	22.9	32.4	38.0
	N	18	18	18	18	18	18
10.0 mg/kg	MEAN	109.3	174.6	246.6	314.2	376.6	416.4
	S.D.	8.3	14.7	19.2	22.7	29.7	36.9
	N	20	20	20	20	20	20
40.0 mg/kg	MEAN	101.4**	166.7**	236.1**	305.1*	365.4	407.3
	S.D.	9.5	14.3	20.8	27.4	33.7	36.6
FEMALE							
	N	20	20	20	20	20	20
CONTROL	MEAN	103.6	151.1	187.0	210.0	235.8	251.1
	S.D.	7.0	10.0	15.2	15.8	29.7	19.3
	N	20	20	20	20	20	20
5.0 mg/kg	MEAN	101.9	147.9	181.5	207.1	228.1	248.5
	S.D.	8.1	13.3	19.3	22.7	25.8	27.5
	N	18	18	18	18	18	18
10.0 mg/kg	MEAN	99.7	148.7	185.5	211.4	237.1	257.6
	S.D.	8.5	11.8	13.3	15.8	19.7	22.2
	N	20	20	20	20	20	20
40.0 mg/kg	MEAN	94.4**	141.3*	177.2	201.9	225.3	241.7
	S.D.	7.0	9.9	12.5	13.9	16.2	16.6

Postnatal development: preweaning observations are summarized in the following sponsor's tables:

Table 7

93-720-28
CP-88,059-1

PRE-WEANING DEVELOPMENTAL INDICES AND BEHAVIOR OF MALE PUPS[a]

DAM NO.		Surface Righting Before culling	Surface Righting After Culling	Appearance of Incisors	Eye Opening	Air Righting	% Failed Visual Cliff
CONTROL	N	20	20	20	20	20	20
	mean	2.8	3.0	10.9	15.1	18.0	6.3%
	s.d.	0.6	0.8	0.7	0.5	0.8	0.1
5.0 mg/kg	N	20	20	20	20	20	20
	mean	2.8	2.9	10.7	14.9	17.7	6.3%
	s.d.	0.9	0.9	1.0	0.5	0.8	0.1
10.0 mg/kg	N	18	18	18	18	18	18
	mean	2.6	2.7	10.8	15.2	17.9	5.6%
	s.d.	0.6	0.7	0.9	0.5	0.9	0.1
40.0 mg/kg	N	20	20	20	20	20	20
	mean	3.0	3.0	11.2	15.6**	18.7**	11.7%
	s.d.	0.7	0.7	0.6	0.5	1.1	0.2

[a]Mean postnatal day of appearance or successful performance, except visual cliff data

**p < .01

Table 8

93-720-28
CP-88,059-1

PRE-WEANING DEVELOPMENTAL INDICES AND BEHAVIOR OF FEMALE PUPS[a]

DAM NO.		Surface Righting Before culling	Surface Righting After Culling	Appearance of Incisors	Eye Opening	Air Righting	% Failed Visual Cliff
CONTROL	N	20	20	20	20	20	20
	mean	3.2	3.5	10.8	14.9	18.0	5.0%
	s.d.	0.7	1.0	0.6	0.5	0.7	0.1
5.0 mg/kg	N	20	20	20	20	20	20
	mean	2.9	3.0	10.8	14.8	17.9	10.5%
	s.d.	0.6	0.9	1.0	0.6	1.0	0.2
10.0 mg/kg	N	18	18	18	18	16	18
	mean	2.7	2.9	10.6	15.0	18.3	9.7%
	s.d.	0.6	0.7	0.5	0.5	0.6	0.2
40.0 mg/kg	N	20	20	20	20	20	20
	mean	3.0	3.1	11.1	15.5**	18.5**	8.5%
	s.d.	0.8	0.9	0.7	0.6	0.9	0.1

[a]Mean postnatal day of appearance or successful performance, except visual cliff data

**p < .01

It wasn't clear what surface righting "before" and "after" culling refers to. The ability for a pup, when placed on its back, to successfully right itself on all four paws was tested daily from Day 1 postpartum until achieved.

Delays (0.5-0.7 days, based on means) in eye opening and air righting were observed in HDM and HDF pups. Ranges (for day of appearance or successful performance) were 14-16 and 16-21 days, respectively, in C pups and 14-17 and

16-22 days, respectively, in HD pups.

Although not statistically significant, the % of pups/litter that failed the visual cliff test was increased in HDM pups, and at all doses in females (not dose-related).

In terms of postweaning developmental landmarks, no drug-related effects were observed on the following: vaginal opening, preputial separation, auditory function test, passive avoidance. straight channel swimming latency, In the Cincinnati Maze, the only notable observation was an increase in the no. of errors during one trial (#9) at all doses in females (1.6, 4.9, 4.6, and 6.9 errors at C, LD, MD, and HD, respectively). A similar, although non-significant, effect was noted during trial #8 (6.4, 10.6, 10.6, and 11.0 errors at C, LD, MD, and HD, respectively). The increase in errors in treated females during trials #8 and 9 resulted in an increase in total errors for Path B37.9, 41.6, 48.8, and 50.1 errors at C, LD, MD, and HD, respectively); however, the differences among grps were not statistically significant. Motor activity was increased in females in a dose-related manner (2-26, 11-27 and 12-55% in LDF, MDF and HDF, respectively) throughout the measurement period (20 min). Overall mean distance traveled was increased by 6, 16, and 19% in LDF, MDF, and HDF, respectively; the increase was statistically significant only at the HD. The greatest differences between C and drug-treated grps were noted at the last measurement interval, suggest some delay in habituation.

In the Functional Observation Battery (FOB), 29 different responses was tested; **only data for the following were summarized**: ease of removal, handling reactivity, rearing, gait, gait score, arousal, defecation, urination, approach response, touch response, click response, tail pinch response, and body temperature. In males, there was a greater reactivity to tail pinch (scores: 1.93 ± 0.37 and 2.23 ± 0.34 for C and HD, respectively) and lowered body temperature (mean difference of -0.32° C) at the HD. In females, there was increased defecation at the MD and HD (0.25 ± 0.47, 0.81 ± 0.91, and 0.73 ± 0.90 for C, MD, and HD, respectively) and a decrease in body temperature at all doses (37.69 ± 0.31, 37.43 ± 0.33, 37.42 ± 0.22, and 37.36 ± 0.23° C at C, LD, MD, and HD, respectively).

Terminal studies

Gross pathology: macroscopic findings were not summarized by the sponsor.

GENOTOXICITY

Ziprasidone was tested in a number of genotoxicity assays. All but one of the studies were submitted in the original IND ( ) and have been reviewed previously (P/T Review, J. J. DeGeorge, Ph.D., 4/13/90). In Dr. DeGeorge's original review, the following genotoxicity studies were reviewed: Ames tests, a mouse lymphoma assay, an *in vitro* cytogenetic assay in human lymphocytes, and *in vivo* cytogenetics assays in mouse. The report submitted in the NDA is a revision of the original report (dated 10/5/89). According to the sponsor, this revision (Study No. 880729-01) contains additional details on methodology, historical control data, and one new assay, an *in vitro* chromosomal aberration assay (conducted in 1995 using more current methodology); no changes have been made to the original data.

Ames test: the genotoxic potential of CP-88,059 was tested in *S. typhimurium* tester strains, TA1535, TA1537, TA98, and TA100 with and without metabolic activation (S9 from rat, mouse). CP-88,059 (0.0002-0.1 mg/plate) and urine (0.05-1.0 mL; 0-30 mg/kg p.o.) from mice treated with CP-88,059 were tested in these strains. One of the following tester strains, sensitive to cross-linking mutagens, was not used either with the drug itself or in mouse urine: *E. coli* WP2uvrA or *E. coli* WP2uvrA(pKM101) or *S. typhimurium* TA102.

In the absence of metabolic activation, increases in the number of revertants were consistently observed at the HC (0.1 mg/plate of CP-88,059) in TA1537. [According to the sponsor, in none of these studies was there any evidence of cytotoxicity or insolubility.] The sponsor's data tables are provided below:

preliminary study:

TABLE 1
CP-88,059-01
BACTERIAL MUTATION ASSAY: SUMMARY OF TEST RESULTS

PLATE INCORPORATION ASSAY WITHOUT S9

Compound	mg/plate	Average Number of Revertant Colonies Per Plate ± S.D.			
		TA 1535	TA 1537	TA 98	TA 100
NONE (DMSO)	---	18 ± 4	16 ± 6	29 ± 4	141 ± 21
CP-88,059-01	0.0002	13 ± 2	18 ± 4	31 ± 7	137 ± 31
	0.001	12 ± 3	13 ± 2	35 ± 5	149 ± 11
	0.005	15 ± 0	17 ± 1	27 ± 5	149 ± 12
	0.02	17 ± 4	18 ± 1	36 ± 4	126 ± 5
	0.1	15 ± 2	39 ± 12	43 ± 5	120 ± 3
Positive Controls					
SODIUM NITRITE	2.0	272 ± 66	---	---	245 ± 51
9-AMINOACRIDINE	0.15	---	1003 ± 383	23 ± 4	---
2-NITROFLUORENE	0.02	---	139 ± 25	1467 ± 47	---
NITROFURANTOIN	0.002	21 ± 6	---	---	1126 ± 295

definitive study, rat:

		TA1537		
		C1	C2	C3
NONE (DMSO)	—	18 ± 4	22 ± 6	18 ± 4
CP-88,059-01	0.0002	19 ± 0	20 ± 4	18 ± 7
	0.001	16 ± 3	22 ± 4	23 ± 7
	0.005	16 ± 1	18 ± 1	26 ± 2
	0.02	21 ± 3	18 ± 6	20 ± 1
	0.1	40 ± 2	24 ± 5	27 ± 5

definitive study, mouse:

		TA1537		
		C1	C2	C3
NONE (DMSO)	—	18 ± 6	25 ± 6	25 ± 6
CP-88,059-01	0.0002	15 ± 3	28 ± 3	31 ± 7
	0.001	17 ± 3	29 ± 4	29 ± 9
	0.005	17 ± 1	24 ± 6	26 ± 6
	0.02	19 ± 8	30 ± 4	33 ± 6
	0.1	51 ± 12	44 ± 10	30 ± 7

key:

C1 ... Direct conditions: Cells + compound

C2 ... Inactive metabolic control: Cells + compound + Rat S9 mixture (minus NADP)

C3 ... Active metabolic conditions: Cells + compound + Rat S9 mixturem (with NADP)

In the preliminary study, individual control (DMSO) replicate values (-S9) ranged from 9-26 revertants. Individual values for TA1537 at the HC were 44, 47, and 25 revertants.

In the definitive rat study, individual control (DMSO) replicate values (-S9) ranged from 14-21 revertants. Individual values for TA1537 at the HC were 42, 38, and 41 revertants.

In the definitive mouse study, individual control (DMSO) replicate values (-S9) ranged from 10-27 revertants. Individual values for TA1537 at the HC were 38, 60, and 55 revertants.

In the preliminary study and in the rat and mouse definitive studies, there was also a slight increase (1.4-1.9 fold) in revertants at the HC with tester strain, TA98, in the absence of metabolic activation.

There were no increases in revertant in any tester strain (±S9) when incubated with mouse urine.

mouse lymphoma (L5178Y/TK+/- locus): the mouse lymphoma assay was repeated 3 times.

The first two assays conducted in the absence of metabolic activation were not used for

assessment of mutagenic potential. In these assays, concentrations of 548-1064 and 271-532 µg/mL were used; "incomplete solubility" was a designated problem at all concentrations. In the 3rd assay (-S9), compound ppt was also noted at all concentrations even though lower concentrations were used (111-399 µg/mL). These data were, however, considered to demonstrate a lack of genotoxic potential for CP-88,059. The data for this assay are presented in the following sponsor's table:

TABLE 2c

CP-88,059-01

MOUSE LYMPHOMA TK$^{+/-}$ ASSAY: SUMMARY OF RESULTS

TEST WITHOUT METABOLIC ACTIVATION

	CYTOTOXICITY				MUTANT SELECTION	
Treatment:	^ Relative % Suspension Growth	Cloning Efficiency **VC	Cloning Efficiency % VC	Total ***Growth	† Mean No. Mutant/10^6 Cells Plated (SD)	Mutant†† Frequency per 10^6 Survivors
Negative Control:DMSO						
1%	100	87	100	100	19(4.0)	22
	100	93	100	100	23(1.5)	25
Test Article: CP-88,059-01 (µg/ml)						
111*	78	78	87	68	17(3.6)	22
134*	74	84	93	69	21(2.9)	25
160*	63	81	90	57	13(2.0)	16
193*	58	76	84	49	12(5.0)	16
231*	56	75	83	46	16(6.0)	21
277*	56	74	82	46	18(2.3)	24
332*	52	61	68	35	18(1.7)	30
399*	50	40	44	22	20(2.8)	50
Positive Control: Ethylmethane sulfonate (µg/ml)						
62†	64	58	64	41	488(15.2)	641
62	104	78	87	90	74(5.8)	95

Abbreviation: SD - standard deviation.

* Incomplete compound solubility in culture medium (1X).

^ The tota daily growth for experimental cultures divided by the average total growth of the solvent controls

** Average viable count (VC) adjusted to a plated cell concentration of 1.5/ml per 33 ml plate.

*** %VC x % relative suspension growth divided by 100.

† mean TFT = average of triplicate plate counts for each test or control concentration.

†† Mutant frequency is calculated by dividing the number of TFT^R colonies (average of triplicate plates) by the number of the cells plated (1 x 10^6) and corrected for the VC for that particular culture.

At the HC, a relative cloning efficiency of 44% and a relative total growth of 22% was obtained; these values do not indicate excessive cytotoxicity. At this concentration, there was an ≈2-fold increase in mutation frequency. [Note: the individual replicate data for this assay could not be found in the submission.]

In the presence of metabolic activation, CP-88,059 was tested at concentrations of 548-1064, 410-798, and 149-532 µg/mL. In the first assay, "excessive" cytotoxicity (defined by the sponsor as a ≤50% total cell growth) and compound ppt occurred at all concentrations. This assay was, therefore, not used for assessment of genotoxic potential. The data from this study were summarized in the sponsor's table below. The relative cloning efficiency and the relative total growth were 55 and 26%, respectively, at the HC. These values are not indicative of unacceptable cytotoxicity. It should be noted that there was a >2-fold increase in mutation frequency at concentrations of 798, 878 and 968 µg/mL.

CP-88,059-01

MOUSE LYMPHOMA TK$^{+/-}$ ASSAY: SUMMARY OF RESULTS
NOT USED TO EVALUATE MUTAGENICITY

TEST WITH METABOLIC ACTIVATION

	CYTOTOXICITY				MUTANT SELECTION	
Treatment	^ Relative % Suspension Growth	Cloning Efficiency **VC	Cloning Efficiency % VC	Total ***Growth	Mean No. Mutant/10^6 Cells Plated (SD)	Mutant Frequency per 10^6 Survivors
Negative Control: DMSO						
1%	100	97	100	100	21 (4.9)	22
	100	88	100	100	19 (2.9)	22
Test Article: CP-88,059-01 (µg/ml)						
548*	40	63	68	27	13 (3.1)	21
601*	62	64	69	43	19 (6.5)	30
660*	55	83	90	50	24 (2.0)	29
729*	53	75	81	43	20 (3.2)	27
798*	62	54	58	36	26 (2.1)	48
878*	62	50	54	44	27 (2.5)	54
968*	68	45	49	32	22 (2.6)	49
1064*	47	51	55	26	12 (3.1)	24
Positive Control: 3-Methylcholanthrene (µg/ml)						
5.37	98	87	94	92	108 (12.1)	124
2.69	106	87	94	100	70 (8.0)	80

In the other assays, there was also compound ppt at all concentrations tested and in one of the assays, the relative cloning efficiency was lower than and the relative total growth was the same as those in the "rejected" study . However, the sponsor used the data from these studies to conclude a lack of a genotoxic response for CP-88,059. The data from these assays are summarized in the following sponsor's tables:

TEST WITH METABOLIC ACTIVATION

Treatment	CYTOTOXICITY: ^ Relative % Suspension Growth	CYTOTOXICITY: Cloning Efficiency **VC	CYTOTOXICITY: Cloning Efficiency % VC	CYTOTOXICITY: Total ***Growth	MUTANT SELECTION: † Mean No. Mutant/10⁶ Cells Plated (SD)	MUTANT SELECTION: Mutant†† Frequency per 10⁶ Survivors
Negative Control: DMSO						
1%	100	117	100	100	22 (5.0)	19
	100	122	100	100	25 (1.5)	20
Test Article: CP-88,059-01 (µg/ml)						
410*	48	82	69	33	22 (5.6)	27
452*	97	75	63	61	24 (4.5)	32
495*	57	80	67	38	22 (3.0)	28
543*	62	71	59	37	25 (2.0)	35
601*	90	60	50	45	26 (4.6)	43
660*	147	44	37	54	20 (1.5)	45
724*	184	32	27	50	8 (4.5)	25
798*	104	30	25	26	13 (3.5)	43
Positive Control: 3-Methylcholarthrene (µg/ml)						
5.37	57	99	83	47	119 (6.2)	120
2.69	102	104	87	89	84 (2.1)	81

REPEAT TEST WITH METABOLIC ACTIVATION

Treatment	CYTOTOXICITY: ^ Relative % Suspension Growth	Cloning Efficiency: **VC	Cloning Efficiency: % VC	CYTOTOXICITY: ***Total Growth	MUTANT SELECTION: † Mean No. Mutant/10^6 Cells Plated (SD)	MUTANT SELECTION: †† Mutant Frequency per 10^6 Survivors
Negative Control: DMSO						
1%	100	108	100	100	19 (4.9)	18
	100	96	100	100	28 (7.6)	29
Test Article: CP-88,059-01 (µg/ml)						
149*	61	80	78	48	19 (3.5)	24
176*	65	67	68	43	23 (1.0)	34
213*	47	92	90	42	20 (3.5)	22
255*	73	69	68	50	19 (2.1)	28
309*	66	91	89	59	31 (1.5)	34
367*	76	70	69	54	13 (6.0)	19
442*	97	59	57	55	18 (4.2)	31
532*	141	57	56	79	13 (4.5)	23
Positive Control: 3-Methylcholanthrene (ug/ml)						
5.37	83	93	91	76	102 (13.2)	110
2.69	86	96	94	83	75 (1.5)	76

The relative cloning efficiency (RCE) was not sufficiently low to be concentration limiting in either assay, although the 25% RCE was probably acceptable. However, compound ppt was a problem at all concentrations. At 601, 660, and 798 µg/mL (but not at 724 µg/mL), there were ≈2-fold increases in mutation frequency. No increases were noted at lower concentrations in the presence of metabolic activation.

No colony sizing was performed on any of the cell cultures.

In vitro cytogenetic assays. the initial study (#88-720-01) was conducted in 1988. In this study, human lymphocytes were incubated with CP-88,059 (solvent: DMSO) for 24 hr in the absence of metabolic activation (phenobarbital-induced rat liver S9) and for 1 hr in the presence of metabolic activation. Mitomycin C (-S9) and cyclophosphamide (+S9) were used as positive controls. A total of 200 metaphases were examined per concentration (100 for positive control). **The data were summarized by the sponsor only for aberrations excluding gaps, i.e., gaps were not included in the overall analyses for any of the assays (±S9).**

In the absence of metabolic activation, unacceptable cytotoxicity (i.e., >95% decrease in MI) was observed at concentrations ≥500 µg/mL. No increase in the no. of cells with chromosomal aberrations was detected at concentrations of 200, 300, and 400 µg/mL. [MI was reduced by ≥50% at all concentrations.] However, the sponsor noted that ppt was present at all of these concentrations. In the presence of metabolic activation, unacceptable cytotoxicity was observed at concentrations ≥200 µg/mL. [The sponsor noted the presence of ppt at all concentrations scored, i.e., 50-150 µg/mL).] The no. of abnormal cells was increased at all concentrations tested (i.e., 50, 100, and 150 µg/mL; statistically significant at the LC and HC). The data are summarized in the following sponsor's table (reproduced in part):

APPENDIX IIIb
CP-88,059-01
HUMAN LYMPHOCYTE ABERRATION ASSAY: INDIVIDUAL DATA VALUES

1 HOUR TREATMENT
WITH METABOLIC ACTIVATION

Treatment	(%) Mitotic Index	No. Cells Analyzed	P	Gaps Ctg	Gaps Csg	Aberrations Ctb	Aberrations Csb	Aberrations R	Aberrations M	No. Abnormal Cells
Negative Control: DMSO										
0.2%	12.6	100	0	0	0	0	0	0	0	0
	13.1	100	0	0	0	0	0	0	0	0
Test Article: CP-88,059-01 (µg/ml)										
50+	7.9	100	0	1	0	2	0	0	0	2
	6.3	100	0	0	0	3	0	1	0	5*
100+	4.6	100	0	0	0	1	0	0	1	2
	5.2	100	0	0	0	0	0	0	0	0
150+	5.7	100	0	1	0	2	1	0	0	2*
	5.8	100	0	1	0	4	0	0	0	4
200+	2.3	--	--	--	--	--	--	--	--	--
	2.0	--	--	--	--	--	--	--	--	--

The mean % of abnormal cells (per 100 metaphases) was 3.5, 1, and 3.0 at 50, 100, and 150 µg/mL, respectively.

A more recent (1995) *in vitro* chromosomal aberration test in human lymphocytes was conducted using more current methodology. In this study, the clastogenic potential of CP-88,059 was tested in the absence and presence of metabolic activation (rat liver S9) using incubation times of 3 and 24 hr (-S9) and 3 hr (+S9). A total of 200 metaphases were examined per concentration (100 for positive controls).

In the absence of metabolic activation, CP-88,059 was tested at concentrations of 320, 400, and 500 µg/mL (3-hr treatment), 205, 256, 320, and 400 µg/mL (24-hr treatment), and 205, 256, 320, and 400 µg/mL (24-hr treatment, repeat). With the 3-hr treatment, the MI was reduced to 64-67% of C at the HC; however, ppt was present at all three concentrations tested. There was no increase in the no. of abnormal cells under these conditions. In the first 24-hr treatment assay, the MI was only reduced to 52-56% of C at the HC; however, ppt was present at all concentrations tested. The number of abnormal cells was increased at 205, 320, and 400 µg/mL. The data are provided in the following sponsor's tables:

TABLE 2b
CP-88,059-01

HUMAN LYMPHOCYTE ABERRATION ASSAY: SUMMARY OF TEST

24 HOUR TREATMENT
WITHOUT METABOLIC ACTIVATION

Treatment[a]	Mean Total Growth Index[b]	Relative Growth[c]	Mean (%) Mitotic Suppression[d]	Total Cells Analyzed	Mean (%) Abnormal Cells	p-Value[e]
Negative Control: DMSO						
1%	1.98	100	0	200	1.0	--
Test Article: CP-88,059-01 (µg/ml)						
164+*	1.22	61	--	--	--	--
205+	1.37	69	45	200	2.0	0.343
256+^	1.59	80	38	200	0.5	> 0.500
320+	1.43	72	40	200	5.5	0.010
400+	1.24	63	48	200	3.0	0.142
500+**	1.62	82	--	--	--	--
Positive Control: Mitomycin-C						
0.05	1.70	86	43	100	18.0	≤ 0.001

a: Two replicate cultures evaluated for each treatment group; when possible 100 cells per culture are analyzed for chromosome damage.

b: Mean Total Growth Index = Mean Day 1 cells/ml / Day 0 Stock Cell Count [3.75 x 10^5 cells/ml].

c: Relative Growth = Total Growth Index of Test Article / Total Growth Index of Negative Control (x) 100.

d: Mean (%) Mitotic Suppression = [One minus the quotient (Mean test Article Mitotic Index / Mean Negative Control Mitotic Index)] (x) 100.

e: Data analyzed by 1-tailed Fisher's Exact test for increases in % abnormal cells compared to control values.

APPENDIX IIId
CP-88,059-01
HUMAN LYMPHOCYTE ABERRATION ASSAY: INDIVIDUAL DATA VALUES

24 HOUR TREATMENT
WITHOUT METABOLIC ACTIVATION

Treatment	Day 1 Cells per ml (x10⁵)[a]	(%) Mitotic Index	No. Cells Analyzed	—P	Gaps Ctg	Gaps Csg	Aberrations Ctb	Csb	R	M	No. Abnormal Cells
Negative Control: DMSO											
1%	7.20	10.0	100	0	1	0	1	0	0	0	1
	7.68	10.8	100	0	2	0	1	0	0	0	1
Test Article: CP-88,059-01 (µg/ml)											
67.2	4.35	--	--	--	--	--	--	--	--	--	--
	5.20	--	--	--	--	--	--	--	--	--	--
84.0	3.91	--	--	--	--	--	--	--	--	--	--
	6.02	--	--	--	--	--	--	--	--	--	--
105+	4.17	--	--	--	--	--	--	--	--	--	--
	4.41	--	--	--	--	--	--	--	--	--	--
131+	4.39	--	--	--	--	--	--	--	--	--	--
	4.20		--	--	--	--	--	--	--	--	--
164+	4.49	--	--	--	--	--	--	--	--	--	--
	4.65	--	--	--	--	--	--	--	--	--	--
205-	4.82	6.2	100	0	3	0	2	0	0	0	2
	5.44	5.1	100	0	1	0	3	3	0	0	2*
256-^	6.70	5.5	100	1	1	0	1	0	0	0	1
	5.29	7.5	100	0	1	1	0	0	0	0	0
320-	5.13	5.5	100	0	2	1	3	3	0	0	6
	5.63	6.9	100	0	1	0	2	4	0	0	5*
400-	4.54	5.6	100	0	2	0	2	3	0	0	5
	4.77	5.2	100	0	2	1	1	0	0	0	1
500-	6.25	--	--	--	--	--	--	--	--	--	--
	5.93	--	--	--	--	--	--	--	--	--	--
Positive Control: Mitomycin-C											
0.05	6.27	5.7	50	0	4	1	7	3	1	0	9*
	6.49	6.2	50	0	4	1	7	2	2	0	9*

a: Day 1 Cells/ml = Mean Coulter Counts (x) Dilution Factor (40).
--: Dashes indicate data not available or determined.
*: Some cells contain more than one aberration.
-: Precipitate was observed in the cultures.
^: Slides were evaluated uncoded after the original three levels.

Abbreviations: Ctg-Chromatid Gap; Csg-Chromosome Gap; P-The number of polyploic cells observed during metaphase collection; Ctb-Chromatid Break; Csb-Chromosome Break; R-Rearrangement; M-Multiple Aberrations (≥ 7).

In the repeat 24-hr assay (-S9), the MI was reduced to ≈58% of C at the HC; however, the sponsor noted the presence of ppt at all concentrations scored. The sponsor noted that the

cultures treated at 256 µg/mL were not analyzed (no reason was found). No increase in the no. of abnormal cells was detected at 205, 320, or 400 µg/mL.

In the first assay in the presence of S9, sufficient cytotoxicity was not achieved (concentration range: µg/mL; 8% reduction in the mitotic index at the HC); therefore, none of the cultures were scored. [The sponsor noted that ppt was observed at all concentrations.] The assay was repeated at the 3 highest concentrations used in the first assay. [It is interesting that the same concentrations were used considering the lack of cytotoxicity achieved in the first assay.] In the repeat assay, in the presence of S9, concentrations of 320, 400, and 500 µg/mL were scored. The MI was reduced to 73% of C at the HC; however, the sponsor noted that a ppt was present at all concentrations. No increases in the % abnormal cells were observed.

The sponsor concluded that the results of this battery of *in vitro* chromosomal aberration tests in human lymphocytes indicate the lack of a clastogenic effect for CP-88,059.

In vivo cytogenetic assays: two *in vivo* chromosomal aberration assays were conducted in CD-1 mice (5/sex/grp). CP-88,059 was administered at single oral (gavage) doses in both assays, 30 mg/kg in the first study (5-6/88)and 200 mg/kg in the second study (3-4/89). Bone marrow was examined for presence of chromosomal aberrations at 6, 24, and 48 hr postdosing; control bone marrow was collected at 24 hr postdosing only. Mitomycin C (3 mg/kg i.p.) was used as the positive control in both studies. **According to the sponsor, concentration of test material was "...verified mathematically rather than analytically; stability in the vehicle was not assessed".**

Approximately 2 hr prior to sacrifice, animals were injected i.p. with colchicine (3 mg/kg). [According to the OECD (1997) guidelines, colchicine (a metaphase arresting agent) should be injected 3-5 hr prior to sacrifice.] At sacrifice, femurs were processed for collection of bone marrow. In neither study was the mitotic index determined. Also, only 50 cells (theoretically in metaphase) were examined per animal. According to the OECD (1194, 1997) guidelines, "The mitotic index should be determined as a measure of cytotoxicity in at least 1000 cells per animal for all treated animals (including positive controls) and untreated negative control animals" and, for scoring of chromosomal aberrations, "At least 100 cells should be analysed for each animal".

In the initial statistical analysis, the results (for both studies) were analyzed using historical instead of concurrent controls. At the request of the Division, the sponsor re-analyzed the data using concurrent controls. Gaps were not scored (or at least data on gaps were not provided) nor was polyploidy.

Study 1: the sponsor noted no clinical signs or death associated with dosing at 30 mg/kg. There were no increases in the relative no. of abnormal cells at 30 mg/kg.

Study 2: the sponsor noted that "decreased activity, decreased respiration and ataxia were observed within 30 minutes after dosing with 200 mg/kg...". These signs were evident within 30 min and persisted for "several hours". There were no drug-related deaths. There was an increase in the relative no. of abnormal cells in males at the 24-hr sampling time (2.4 ± 2.6 vs 1.6 ± 1.7 in VC) and in females at the 6-hr sampling time (2.0 ± 2.4 vs 1.2 ± 1.8 in VC). Neither of thesd differences achieved statistical significance.

Mass balance (% of total radioactivity)

STUDY NO.	SPECIES/STRAIN	SEX	DOSE (ROUTE)	URINE (%)	FECES (%)	BILE (%)	TOTAL (%)
DM-93-128-8	CD-1 mouse	M	10 mg/kg p.o.	33.40-32.53	74.88/72.23		108.28/104.76
		F		32.57/31.45	74.45/72.35		107.02/103.80
DM-93-128-11	Sprague-Dawley rat (intact)	M/F		18.07	92.53	---	110.6
	Sprague-Dawley rat (bile-cannulated)			18.73	48.80	34.64	99.00
DM-93-128-11	Long-Evans rat (bile-cannulated)	M		22.46/21.89	63.38/62.21	16.20/14.68	102.29/98.54
		F		19.60/18.63	49.77/51.62	21.19/19.88	90.55/90.13
DM-94-128-16	Beagle dog (bile-cannulated)	M	5 mg/kg p.o.	5.9-6.0	86.3-87.5	8.2-9.3	
		F		15.3-15.4	79.7-80.1	8.6-9.0	

Plasma metabolites (% of radioactivity)

METABOLITE	C D-1 MOUSE	LONG-EVANS RAT*	BEAGLE DOG
M1	detected	detected	1.1-1.2
M2	detected	detected	1-4
M3	detected	detected	--
M4	detected	detected	2-5
M5	---	detected	3-4
M5a	---	detected	
M6	detected	detected	
M7	≥10 (F)	---	6-8
M8 (sulfone)	32-34	---	10-20
M9	14-20	detected	25-30
M10 (sulfoxide)		detected	
M13 (parent)	7-12	detected	15-20

* 70-84% of total plasma radioactivity was accounted for in rat

respectively.

General pharmacology

The effects of ziprasidone in isolated guinea pig tissue preparations (aorta, ileum, atria) and in rat uterus were tested. Ziprasidone inhibited NE-induced contractions in aorta and H_1-induced contractions in ileum, but had no effect in the other paradigms.

In the Irwin screen conducted in CD-1 mice (3/grp), ziprasidone (acute dosing) exhibited the following effects: reduced respiration (3.2-1000 mg/kg p.o.), ptosis (≥3.2 mg/kg p.o.), inverted screen defects (≥10 mg/kg p.o.), miosis, decreased urination, decreased spontaneous motor activity, and loss of tail pinch response at ≥32 mg/kg p.o., and complete immobility (i.e., catalepsy) in all animals at 320 and 1000 mg/kg.

Safety pharmacology

Ziprasidone's effects on cardiovascular parameters, acid-base balance, renal excretion, GI transit time, and gastric acid secretion were tested in a series of studies.

Cardiovascular effects: assessment of ziprasidone's effect on cardiovascular parameters was minimal. One study was conducted in 5 conscious Mongrel dogs; however, 2 of the dogs vomited after dosing and were not replaced. Therefore, ziprasidone's effects on hr, MAP, and QA-interval were assessed only in 3 dogs following an acute 6 mg/kg p.o. dose. In these dogs, ziprasidone increased heart rate (≤60%) from 45 min postdosing on, but no effects were noted on MAP or QA-interval. Plasma levels in these dogs ranged from
postdosing.

Other effects: ziprasidone had little or no effects on acid-base balance (3 drug-treated rats, single dose of 12 mg/kg p.o.), renal excretion (rats, "n" not specified in report, single doses of 1.6, 6, and 12 mg/kg p.o.), or GI transit (rats, 8/grp; single doses of 1.6 and 6.0 mg/kg p.o.). Ziprasidone did reduce (65-58%) gastric acid secretion in pylorus-ligated rats (10/grp) at doses of 1.6 and 6.0 mg/kg p.o.; however, the effect was not dose-related.

PK/ADME

The PK/ADME of ziprasidone was evaluated primarily in CD-1 mice, rats (Sprague-Dawley, Long-Evans), and Beagle dog. The PK data collected in these animals are summarized in the attached table. Of the total radioactivity in plasma, the parent compound accounted for 15-20% (C_{max}) and 8-9% (AUC) in mouse (10 mg/kg p.o.), 50-60% (C_{max}) and 40-50% (AUC) in Long-Evans rat (10 mg/kg p.o.), and 28-34% (C_{max}) and 12-21% (AUC) in Beagle dog (5 mg/kg p.o.). In humans, the parent compound accounted for 52-56% of total plasma radioactivity following a single 20-mg oral dose. The primary route of elimination was fecal in all three species. In bile-cannulated rats and dogs, bile radioactivity accounted for 20-35 and 8-9% of dose radioactivity, respectively.

Tissue distribution studies were conducted in Sprague-Dawley and Long-Evans rats following single oral doses (5, 10 mg/kg). In Long-Evans rats, the highest levels of radioactivity were detected in liver, followed by eye and kidney; lowest levels were detected in brain. Peak levels were achieved at ≈2 hr, except in eye. At 72 hr postdosing, 65-70% of peak levels were still detectable in eye. Retention of radioactivity was assessed in selected tissues in Long-Evans and Sprague-Dawley rats. In the pigmented rats, detectable levels of radioactivity were still detectable in eye and testes 6-mo to 1-yr following dosing (20-26 and 12-10% of peak levels, respectively). In non-pigmented rats, long-term retention of radioactivity was detectable only in testes (10% of peak levels). In pigmented eye, the highest levels of radioactivity were

PK of parent compound (ziprasidone)

STUDY NO.	SPECIES/ STRAIN	SEX	DOSE (mg/kg)	ROUTE	C_{max} (ng/mL)	T_{max} (hr)	$t_{1/2}$ (hr)	$AUC_{(0-\infty)}$ (ng•hr/mL)	Cl (mL/min/kg)	V_d (L/kg)	F (%)
DM-94-128-1	CD-1 mouse	M	5	i.v.			0.41	1237	67.4	1.9	
			10	p.o.	221	1		678			27
			100	p.o.	917	1		6093			25
		F	5	i.v.			0.53	1356	61.5	2.4	
			10	p.o.	441	1		2411			89
			100	p.o.	1663	1		15905			59
DM-93-128-10		M	10	p.o.	269			605			
		F			302			708			
DM-93-128-2	Long-Evans rat	M	1	i.v.			0.91	734 ± 284	25 ± 9	1.4 ± 1.1	
			25	p.o.	1753 ± 1346	0.5 ± 0.0		4920 ± 881			27 ± 5
		F	1	i.v.			1.1	1215 ± 132	14 ± 2	0.7 ± 0.2	
			25	p.o.	1500 ± 1057	0.6 ± 0.4		6837 ± 1664			22 ± 6
DM-93-128-14		M	10	p.o.	982			2546			
		F			1435			4429			
DM-93-129-4	Beagle dog	M	0.5	i.v.			2.3	535 ± 49	15.7 ± 1.4	2.4 ± 0.2	
			2	p.o.	139 ± 50	1.5 ± 0.58	2.8	752 ± 147			36 ± 9
DM-94-128-16	Beagle dog	M	5	p.o.	132			428*			
		F			277			2008*			

*0-24 hr

PHARMACOLOGY

Pharmacology related to mechanism of action/side effect liability

In vitro: the *in vitro* binding affinity for dopamine (D_1, D_2, D_3, and D_4), serotonin ($5HT_{2A}$, $5HT_{1A}$, $5HT_{2C}$, and $5HT_{1D}$), α-adrenoceptors (1, 2), and the histamine H_1 receptors were assayed. Ziprasidone exhibited high affinity for the D_2, D_3, $5HT_{2A}$, $5HT_{2C}$, $5HT_{1A}$, and $5HT_{1D}$ receptors (K_i's of 4.8, 7.2, 0.4, 1.3, 3.4, and 2 nM, respectively). Fairly high affinity was also exhibited for the α_1 receptor (K_i = 10 nM). Ziprasidone exhibited an ≈12-fold greater affinity for the $5HT_{2A}$ receptor as compared to the D_2 receptor. According to the sponsor's data, risperidone had a 7-fold higher affinity and haloperidol had a 64-fold lower affinity for the $5HT_2$ as compared to the D_2 receptor.

In *in vitro* functional tests, ziprasidone exhibited antagonist effects at the D_1, D_2, $5HT_{2A}$, $5HT_{2C}$, $5HT_{1D}$, and α_1-receptor, but agonist effects at the $5HT_{1A}$ receptor (80% of the activity of a full agonist).

Ziprasidone exhibited moderate affinity for the NE and 5HT reuptake sites (K_i ≈ 50 nM) and low affinity for the DA uptake site (K_i = 260 nM). In functional studies, ziprasidone inhibited reuptake of NE, 5HT, and DA in rat brain crude synaptosomal preparations, but was more potent at the NE and 5HT sites. Ziprasidone did not inhibit MAO activity in rat brain or liver homogenates.

In vivo: ziprasidone's effect on DA and 5HT turnover in forebrain and/or striatum was tested in male Sprague-Dawley rats in two separate experiments. At doses of 0.32-10 mg/kg p.o., ziprasidone increased DA turnover in forebrain, resulting in dose-dependent increases in DOPAC and HVA. DA levels and 5HT levels and turnover were not affected. At doses of 1-10 mg/kg p.o., ziprasidone increased dopamine release in both frontal cortex and striatum; however, the effect in cortex was dose-related (maximum increase 200%) whereas that in striatum was noted only at the HD (40% increase).

Ziprasidone was tested in a number of behavioral paradigms in male Sprague-Dawley rats designed to assess both efficacy and side-effect liability. Ziprasidone exhibited effects in all paradigms tested. Inhibition of amphetamine-induced locomotor activity and the conditioned avoidance response have predictive validity for clinical efficacy. In these paradigms, ziprasidone exhibited effects at ID_{50}'s of 1.5 (range:) and 2.6 (range:) mg/kg p.o. Unfortunately, in the CAR paradigm, escape behavior was not tested; therefore, drug-induced changes in sensitivity to the electric-shock could not be assessed. Ziprasidone also exhibited activity in paradigms considered to predict side-effect liability, however, the effective doses varied to a greater extent. Ziprasidone inhibited apomorphine-induced stereotypy at an ID_{50} similar to that in the efficacy paradigms (i.e., ID_{50} = 2.4 (range:) mg/kg p.o.; however, the $EC_{20\ sec}$ for inducing catalepsy was 12 (≈10-15 mg/kg). Spontaneous motor activity was inhibited [ID_{50} = ≈9 (range:) mg/kg].

Activity of metabolites: the *in vitro* binding affinity of the sulfoxide (M10) and the sulfone metabolite were tested for the D_2, D_3, D_4, and the $5HT_{2A}$ receptors. The sulfone exhibited no affinity (i.e., $K_i \geq 1$ μM) for any of the receptors assayed. The sulfoxide exhibited weak to very-weak affinity for the D_2 and the $5HT_{2A}$ receptors (K_i = 780 and 150 nM, respectively). For comparison, ziprasidone exhibited 200- and ≈375-fold greater affinity for these receptors,

detected in the ciliary body, with slightly lower levels noted in iris. Radioactivity was low, but detectable, in choroid and uvea. No radioactivity was detectable in aqueous or vitreous humor. This pattern of binding in eye is not entirely consistent with melanin distribution in eye. For example, the choroid contains a significant amount of melanin, but only low levels of radioactivity were detected in this area. In binding experiments using synthetic melanin and bovine retina homogenates, >90% of added ziprasidone bound to both sources of melanin (with two different binding sites). Washing with 50% ethanol resulted in the removal of 98% of bound ziprasidone; however, changes in ionic strength of the medium had no effect on binding. The sponsor noted that these data would suggest a hydrophobic interaction between ziprasidone and melanin.

Metabolism studies of ziprasidone were conducted in mouse, rat, dog, and human. The major metabolic pathways shared by these species were as follows:

(1) N-dealkylation of the ethyl side chain at the piperazinyl nitrogen, resulting in cleavage products.

(2) oxidation on the benzisothiazole moiety, at other than the sulfur atom, resulting in M6 and M9.

Other pathways shown to be major in some species and minor in others were as follows:

(1) oxidation at the sulfur atom, resulting in formation of the sulfoxide (M10) and the sulfone (M8) [major in mouse, rat, dog]

(2) hydroxylation on the 3-position and subsequent oxidation of the benzisothiazole moiety, resulting in M4A and M7 [major in rat, dog, human]

Parent compound and identified metabolites accounted for ≈80, 70-84, 70-83, and 58-70% of total plasma/serum radioactivity following oral dosing in mouse, rat, dog, and human, respectively.

Involvement of cytochrome P450 enzymes in the *in vitro* metabolism of ziprasidone was assessed using human liver microsomes. The two major pathways identified *in vitro* were (1) oxidation at the sulfur atom of benzisothiazole and (2) oxidation on the piperazine ring. The three metabolites formed were the sulfoxide, the sulfone, and the desethylene metabolite, ethylenediamine. The ethylenediamine metabolite was not identified in *in vivo* human studies. Ketoconazole added to the incubation mixture inhibited the formation of the sulfoxide and sulfone metabolites by 77%, indicating the involvement of CYP3A4 in the formation of these metabolites. In further inhibition studies, ziprasidone was shown to inhibit CYP2D6 and CYP3A4 *in vitro* (K_i = 5.4 ± 4.8 and 64 ± 22 µM, respectively), but not CYP1A2, CYP2C9, or CYP2C19.

In binding experiments, ziprasidone was to extensive bind to human albumin and α_1-acid glycoprotein (≈98%). No interactions were noted between ziprasidone and either warfarin or propranolol. In a separate study, protein binding in plasma of cebus and cynomolgus monkeys and human was determined to be >99% following i.m. administration in monkey (0.5 or 1.5 mg/kg; there was a discrepancy in the report) and oral administration in humans (80 mg b.i.d.). It should be noted that the n's were minimal, and that methodological difficulties (not further specified) resulted in the loss of data from 4/5 dialysis cells for monkey plasma.

TOXICOLOGY

Acute studies

Acute toxicity studies were conducted in albino mice (500, 200 mg/kg p.o., 300, 500, 1000 mg/kg i.p.) and Sprague-Dawley rats (500, 2000 mg/kg p.o., i.p.). Drug-related deaths occurred only in male mice (100 mg/kg i.p.). Sedation was the primary clinical sign, and appeared to have a more rapid onset and a longer duration with i.p. dosing. No target organ for toxicity was identified. LD_{50}'s were calculated as follows: >2000 mg/kg p.o. (mice, rats), >2000 mg/kg i.p. (rats), 500-1000 mg/kg i.p. (mice).

Subchronic studies

Subchronic oral toxicity studies were conducted in Long-Evans rats (2-wk, 1-mo) and Beagle dogs (2-wk, 1-mo).

In rat, ziprasidone was administered by gavage at doses of 0, 5, 25, and 25 mg/kg in the 2-wk study, and 0, 10, 40, and 160 mg/kg in the 1-mo study. The 2-wk study was inadequate due to the small number of animals (3/sex/grp) and the limited histopathology. The report for the 1-mo study was missing a complete summary of histopathology findings. There were no drug-related deaths in either study. Sedation was noted at all doses in both studies. Body weight was reduced compared to controls at the MD and HD (6-15%) in the 2-wk study and **at all doses (10-25%) in the 1-mo study**. No drug-related effects were noted on hematology/ urinalysis, ophthalmology (1-mo study), gross pathology, or histopathology in either study. A complete battery of organ were not weighed in the 1-mo study. No changes in liver, kidney, or testis wt were detected except those related to changes in body weight.

In dog, ziprasidone was administered by gavage at doses of 0, 2, 5, 10, and 20 mg/kg in the 2-wk study and 0, 10, 20, and 40 mg/kg in the 1-mo study. Dosing was once daily in the 2-wk study and b.i.d. in the 1-mo study (i.e., 0, 5, 10, and 20 mg/kg b.i.d.). The 2-wk study was inadequate due to the small number of animals tested (1/sex/grp) and the lack of terminal studies. The number of animals used in the 1-mo study was minimal (3/sex/grp). There were no drug-related deaths in either study. Sedation was the primary clinical sign, with severity being dose-related. In the 2-wk study, other clinical signs were noted (e.g., splayed hindlimbs, intermittent tremors, ptosis, and shallow breathing). No drug-related effects were observed on body weight, food consumption, ECG (no data provided), hematology, urinalysis, or histopathology. Increases in ALT and AST were detected at the MD and HD in the 1-mo study. As in rat, a complete battery of organ weights were not collected; only kidney, liver, and testes were weighed. No drug-related effects on the weight of these organs were noted.

Chronic studies

Chronic oral toxicity studies were conducted in Sprague-Dawley rat (6-mo) and Beagle dog (6-mo, 1-yr).

In rat, ziprasidone was administered by gavage at doses of 0, 10, 40, and 200 mg/kg. There were 11 unscheduled deaths, 8 of which were attributed to trauma during dosing or during recapture. [The nature of the injuries attributed to trauma sustained during re-capture suggested the possibility at the personnel responsible for care and/or testing of the animals were not sufficiently trained. This issue was referred to DSI, but was not adequately resolved.] The other three deaths were not dose-related. Sedation was the primary drug-related clinical sign and was observed at all doses, with severity being dose-related. Body weight was affected in both males and females; however, males were more markedly affected than females. Final body weights were reduced (compared to CM) by 17, 31, and 35% in LDM, MDM, and HDM. In females, final mean body weights (compared to CF) were slightly elevated in LDF, but reduced at the MD and HD (6 and 9%, respectively). No marked drug-related effects were observed on ophthalmology, hematology, clinical chemistry, or urinalysis parameters. There were small changes in wbc ct and LFTs at the HD; however, the effects were small (LFT increased by <2-fold). A complete battery of organ weights were not obtained;

only kidney, liver, and testes were weighed. Changes in the weight of these organs were consistent with those in body weight. According to the sponsor, there were no gross findings; however, no summary table(s) or line listings were provided. Microscopic findings were detected in lung, adrenal gland, and prostate. Lung changes consisted of pleuritis in a few HD animals and multifocal granulomatous pneumonia. The latter finding was observed in all grps, including controls; however, the incidence and severity of the finding was dose-related. The granulomas were characterized as accumulations of foamy macrophages; the sponsor attributed this finding to "chronic, low level aspiration of the compound during dosing". However, there was no mention of detection of drug-related particles in lung samples. Foamy macrophages can result from drug-related phospholipidosis, as a reaction to inhalation of foreign bodies, or the cause may not be evident. The information provided was not sufficient to determine the possible cause. However, considering the dose-related incidence in males and females, a direct drug-related effect cannot be dismissed. Adrenal gland changes consisted of multifocal cystic degeneration and telangiectasia in MDF and HDF, and diffuse hypertrophy in MDF and HD animals. The diffuse hypertrophy was characterized as increased cytoplasmic mass in cells of the zona fasciculata. The sponsor attributed this finding to stress, and the blood vessel changes and cystic degeneration to age-related degeneration. However, these explanations are not consistent with the dose-related incidences of these findings. The sponsor attributed the prostatitis to age and/or stress, as well as to "...hormones and immunologic factors..." Again, the dose-related severity and incidence would suggest some drug involvement.

In dog, ziprasidone was administered at doses of 0, 5, 10, and 40 mg/kg p.o. (gavage; MD and HD given as 5 and 20 mg/kg b.i.d.) in the 6-mo study and at doses of 0, 5, 10, and 20 mg/kg p.o. (gavage); MD and HD given as 5 and 10 mg/kg b.i.d.) in the 1-yr study. In both studies, the "n" was 4/sex/grp. There was only one unscheduled death; 1 HDM was sacrificed moribund in the 6-mo study. This HDM was sacrificed after fracturing several teeth and the jaw during cage-biting; the sponsor considered this death drug-related. Drug-related clinical signs (e.g., reduced motor activity, recumbency, leaning/pressing against cage, pawing, unusual posture, tremors, hypersalivation , ptosis, aggressive behavior, pacing/circling, muscle fasciculation) were evident at all doses in both studies. Except for clinical signs and sporadic increases in ALT (2-3 fold) in individual animals (1 CM, 2 LDM, 3 HDM, and 2 HDF), no other potentially drug-related effects were noted in the 1-yr study [i.e., on body wt, food consumption, physical/vital signs, ECG, blood pressure, ophthalmology, hematology, clinical chemistry, urinalysis, terminal studies (selected organ wts, gross- and histo-pathology)]. In the 6-mo study, a number of additional findings were noted. Body wt gain was reduced at all doses in males (total body wt gain: 26, 16, 9, and -2% in CM, LDM, MDM, and HDM, respectively); no effects were observed in females. Food consumption did not appear to be affected in either males or females. In addition, no drug-related effects were observed on ECG, SAP, rectal temperature, hematology, or urinalysis parameters. Increases in alkaline phosphatase (4/8 HD: 243-404 U/L) and ALT (8/8 HD: 58-183 U/L) were noted at the HD. No drug-related effects (separate from those on body wt) were noted on kidney, liver, or testis wts (the only organs weighed). No gross pathology findings were noted, except for the fractured teeth and maxilla in the HDM that was sacrificed. Histopathology findings were noted in kidney (lipofuscin deposits in renal proximal tubules in MD and HD animals) and liver (intrahepatic cholestasis in HD animals, characterized by the sponsor as "... bile plugs within the canaliculi and phagocytized bile within Kupffer cells...". The liver findings were consistent with the clinical chemistry findings.

Cardiovascular effects:

Cardiovascular effects were not observed in either the 6-mo or 1-yr studies in dog. In the 6-mo study, ECG measurements (PR, QRS, QT intervals) were quantitated, according to the sponsor, "...from one cardiac cycle in lead II" twice prior to the start of the dosing period and on Days 43-47, 92-95, and 168-169 of dosing. [Seven leads were recorded (i.e., I, II, III, aVR,

aVL, aVF, CV_6LL), but only lead II was used for calculating ECG parameters.]

In the 1-yr study, ECG recordings were obtained using leads I, II, III, aVR, aVL, aVF, and CV_6LL twice prior to the start of the dosing period, and on Days 92, 183, 274, and 351 of dosing (at ≈2 hr postdosing). The lead(s) used to calculated ECG parameters was not specified, nor were the data provided.

CARCINOGENICITY

Two-year carcinogenicity studies were conducted in Swiss mice and Long-Evans rats.

Dose-selection for the definitive studies were based on 1- and 3-mo dietary studies in mice and 3-wk and 3-mo dietary studies in rat (only the 3-mo study was conducted in Long-Evans rats, the strain used in the 2-yr study).

Mouse: CP-88,059 was administered orally in the diet to 50/sex/grp at doses of 0, 0, 50, 100, and 200 mg/kg. Dosing was initiated at 50 mg/kg for the 3 drug-treated grps, and increased after 14 days to 100 mg/kg in the MD and HD grps, then to 200 mg/kg in the HD on Day 29. Dosing was continuous for 720-724 days. Observations consisted of the following: clinical signs, body wt, food/water consumption, ophthalmology, and terminal studies [organ wts (brain, heart, kidneys, liver, testes only), gross and histo-pathology]. In addition, pituitary sections from selected animals were examined using immunocytochemistry in order to better characterize pituitary tumors in those animals.

There were no significant differences in survival rate among grps. According to the sponsor, there were also no drug-related clinical signs (data were not summarized) or on ophthalmology (anterior segment of eye) examination. Body wt was reduced in HDM and HDF (8 and 10%, respectively) as compared to C grps. Body wt gain was reduced in all grps by the end of the dosing period (8, 19, and 34% in LDM, MDM, and HDM, respectively, and 10, 33, and 31% in LDF, MDF, and HDF, respectively). In females, body wt was increased (compared to CF) at the LD up to Day 512 and sporadically at the MD up to Day 337. Decreases in body wt relative to CF were noted only toward the end of the dosing period, even at the HD. Food consumption was reduced in all drug-treated grps (5-13%).

The gross pathology findings were not summarized, except for selected findings considered "noteworthy" by the sponsor. Of these, only pituitary and skin findings were increased in drug-treated grps (females only). Pituitary masses were observed at all doses in females (4-5/50) and the incidence of pituitary enlargement was increased in MDF and HDF. The incidence of skin masses was elevated at all doses in females, but was highest in HDF. A complete battery of organ/tissue wts was not performed. The wts of selected organs were not affected in females. In males, absolute and relative wts of all organs analyzed, except for brain, were reduced primarily at the HD. Absolute, but not relative, brain wt was reduced (LDM, HDM). Microscopic findings were observed primarily in females. Non-neoplastic findings in females consisted of the following: (1) cervical node plasmacytosis (adrenal) (HDF), (2) hypersecretion of the Harderian gland (all doses), (3) foam cell foci in lung (HDF), (4) ovarian atrophy (MDF, HDF), (5) pituitary hyperplasia (all doses), (6) mammary gland mononuclear cell infiltrates (HDF), galactocele (all doses), and hyperplasia (all doses), (7) mononuclear cell infiltrates in lung (all doses), and (8) uterine inactivity (all doses). Neoplastic findings in females consisted of the following: (1) bronchiolar-alveolar adenoma of the lung (HDF), (2) pituitary gland adenoma (all doses, most involving the pars distalis) and carcinoma (HDF), and (3) mammary gland adenocarcinoma (all doses). An examination of the individual data indicated that changes (non-neoplastic/neoplastic) in both mammary gland and pituitary gland were detected in 7/50 LDF, 19/50 MDF, and 29/50 HDF. In terms of neoplasms, 2 LDF, 2 MDF, and 10 HDF had tumors in both tissues. That is, tumors of both the pituitary and mammary glands were more often seen in the same animal at the HD than at the lower

doses; however, changes in these two tissues were not particularly well correlated. In 20/50 LDF, 14/50 MDF, and 14/50 HDF, changes were observed in either mammary gland or pituitary gland. In males, the following non-neoplastic findings were detected: (1) slight increase in hyperplasia of the spindle cells (MDM, HDM) and accessory cortical tissue (HDM) in adrenal gland, (2) hypersecretion of the Harderian gland (all doses), (3) foam cell foci (MDM, HDM), and (4) mononuclear cell infiltrate in thyroid (HDM). Neoplastic findings in males were limited to the Harderian gland (adenoma) (MDM, HDM).

Statistical Evaluation: according to the sponsor's analyses, the only statistically significant findings were as follows: (1) ovarian atrophy (HDF), (2) pituitary gland hyperplasia and adenoma (pars distalis) (HDF), (3) mammary gland galactocele (HDF), hyperplasia (HDF), and adenocarcinomas (HDF), and (4) uterine inactivity (HDF). Except for the lung findings, all differences were significant at $p = 0.0001$ level (trend test). The lung findings in HDF were significant at $p = 0.04$ (trend test).

According to the Statistician's review (Roswitha Kelly, HFD-710; cf. Addendum B), the following neoplastic findings achieved statistical significance in female mice: (1) adenoma of the pituitary gland, (2) adenoma and carcinoma combined of the pituitary gland, (3) adenocarcinoma (incidental and fatal) of the mammary gland. Pairwise comparisons of C and HD grps indicated that differences between these groups for tumors of the pituitary and mammary glands were highly statistically significant. No statistically significant findings were identified in male mice.

At this reviewer's request, the statistician conducted pair-wise comparisons of the lower dose grps with the control grps. The results of these additional statistical tests were as follows: (1) all pair-wise comparisons with controls were highly significant for pituitary adenoma, (2) with pituitary adenoma + carcinoma, the results were not further evaluated because carcinomas were detected only in the HD grp. (3) all pair-wise comparisons with controls were significant for mammary gland adenocarcinomas.

Conclusions: The adequacy of the doses was based solely on the drug-related effects on body weight in HDM and HDF (8 and 10%, respectively). In this reviewer's opinion, it is unfortunate when body weight effects alone define an MTD, since the primary reason for maintaining body wt is to avoid presumed protective effects of reduced body wt gain (i.e., a decrease in the spontaneous rate of tumors). There is also the possibility that, without associated toxicities, the reductions in body wt may reflect only unpalatability of the diet (no gavage dosing studies were available for comparison). This is particularly relevant for male mice, in which no other drug-related toxicities were observed. Neoplastic findings (pituitary adenoma, adenoma/carcinoma combined; mammary gland adenocarcinoma) were observed in females at all doses. The microscopic changes in ovary, pituitary, and mammary gland are consistent with elevations in prolactin.

Drug-effects on serum prolactin were not tested in the 2-yr carcinogenicity; however, a special study was conducted after completion of the 2-yr study to assess drug-related changes in serum prolactin. In this special 1-mo dietary study in CD-1 mice, ziprasidone was reported to increase serum prolactin levels in females, but not males, at all doses tested (50, 100, and 200 mg/kg). Only the effects at the MD and HD were statistically significant, and increases were not dose-proportional (≈4-fold increases at both doses). [In general, there was marked interanimal variability in serum prolactin levels.] It should be noted, however, that drug-related effects were, in general, greater in the 1-mo study than in the 2-yr carcinogenicity study. For example, there were 3 drug-related deaths in HDF and clinical signs (decreased activity) at all doses; no drug-related mortality or clinical signs were observed in the 2-yr study at the same doses. [It is possible that the dose-escalation procedure in the 2-yr study (i.e., increases in dose from 50 mg/kg to final MD and HD over 15-29 days) prevented mortality, but this would not explain the drug-related decreases in activity observed at the LD in the 1-mo study. According to the sponsor, doses were gradually increased in the dose-selection and 2-

yr studies to "...attenuate the expected initial decrease in body weight..." No mention was made of more serious toxicities.] In addition, body wt effects were similar in the 1-mo and 2-yr studies. It is unfortunate that TK data were not available for either study.

Rat: CP-88,059 was administered orally in the diet to Long-Evans rats at doses of 0, 0, 2, 6, and 12 mg/kg. Doses were titrated to the final levels over the first 2-4 wks of the study. Observations consisted of the following: clinical signs, body wt, food consumption, ophthalmology, hematology, clinical chemistry, urinalysis, TK, and terminal studies [organ/tissue wts (kidney, liver, brain, heart, adrenals, and testes), gross pathology, histopathology]. The control grps were combined for statistical analysis.

There were no drug-related increases in mortality; the tendency was for survival rate to be higher in HD grps. According to the sponsor, there were no drug-related clinical signs or findings on ophthalmology examination. Body wt was reduced primarily in HDM (9-10%) and HDF (19%) compared to C grps. The effect was most prolonged in males, being evident from Day 57 through Day 568. By the end of the dosing period, body wts were fairly similar among grps due to increased body wt loss in CM. In females, body wt was reduced from Day 400 on at the HD. Overall body wt gain was reduced only in HDF (22%). Food consumption tended to be lower throughout most of the dosing period in HDM and HDF. In HDM, however, food consumption was somewhat increased compared to CM from Day 596 on. There were no apparent drug-related effects on hematology parameters. Clinical chemistry findings were not marked, consisting of decreases in total protein and albumin in males (7-10%, all doses) and slight decreases in Na in HD animals (2%).

There were no drug-related macroscopic findings. Changes in organ/tissue wts were consistent with drug-related effects on body wt. Microscopic findings were noted in skin (ß-kerato-acanthoma) in HDM and in mammary gland (fibroadenoma) in HDF. There was also a slight increase in corneal mineralization in HDM and ß-thecal/granulosa cell tumor of the ovaries were detected only in 2/50 HDF.

Plasma levels of CP-88,059 were collected during Days 197-198 of dosing. C_{max} was achieved ≈6 hrs after the start of the dark cycle. Plasma exposure, based on both C_{max} and AUC, was ≈2-fold higher in females than in males at all doses. [Unfortunately, estimates of variability were not provided for AUC data.] Both C_{max} and AUC increased fairly linearly with dose, with only slight decreases (8-12%) in dose-corrected exposure with increasing dose. The data are reproduced below (C_{max} 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

Statistical evaluation: according to the sponsor's analyses, the increase in ß-kerato-acanthomas in HDM and in mammary gland fibroadenomas in HDF were the only statistically significant findings. The p-values (trend tests) for these findings were 0.0079 and 0.049, respectively. However, the finding in males was no longer statistically significant when adjustment of multiple pair-wise comparisons was made.

According to the Statistician's review (Roswitha Kelly, HFD-710), none of the neoplastic