in fetuses at 200 or more mg/kg/day (Table 3.5.3.4). There were no fetal skeletal malformations and there was no significant difference in the frequency of skeletal variations among the groups. There was a significant ($p < 0.01$) decrease in the number of ossified caudal vertebrae in the 1000 mg/kg/day group (12% reduction relative to control).

TABLE 3.5.3.2
FOOD CONSUMPTION IN RATS TREATED ORALLY WITH OLMESARTAN MEDOXOMIL

Dose (mg/kg)	Number of pregnant females	Food intake (g) at each gestational day							Number of nursing dams	Food intake (g) at each day after delivery			
		7	9	11	13	15	17	20		4	8	15	22
Control	38	27.3 ±0.54	28.2 ±0.46	28.3 ±0.48	28.4 ±0.42	27.6 ±0.67	30.0 ±0.53	28.9 ±0.47	13	48.8 ±1.61	55.3 ±1.07	62.7 ±1.38	85.0 ±1.77
40	39	27.1 ±0.44 P=0.7630	27.2 ±0.39 P=0.1164	27.1 ±0.49 P=0.0682	26.7* ±0.41 P=0.0042	26.3 ±0.55 P=0.1246	28.5 ±0.45 P=0.0370	27.9 ±0.49 P=0.1264	13	48.5 ±1.75 P=0.8980	55.2 ±1.30 P=0.9640	64.1 ±1.01 P=0.4268	80.2 ±1.71 P=0.0644
200	38	26.2 ±0.33 P=0.1021	25.7* ±0.30 P=0.0001	24.8* ±0.33 P→0.0000	25.4* ±0.41 P→0.0000	23.6* ±0.47 P→0.0000	24.7* ±0.52 P→0.0000	25.4* ±1.03 P=0.0031	13	44.0[a] ±4.06 P=0.2935	50.5 ±3.95 P=0.2611	58.9[b] ±1.43 P=0.0716	80.8 ±1.00 P=0.0538
1000	33	27.4 ±0.38 P=0.8096	26.8 ±0.42 P=0.0413	25.6* ±0.39 P=0.0001	25.1* ±0.63 P→0.0000	23.0* ±0.52 P→0.0000	24.6* ±0.42 P→0.0000	21.4* ±1.55 P→0.0000	9	47.1[c] ±5.10 P=0.7697	55.4 ±1.94 P=0.9531	63.7 ±2.90 P=0.7215	84.7 ±3.97 P=0.9402

Data are expressed as mean S.E.M. * Significant at 1% level compared with control
a) mean of 12 dams, b) mean of 11 dams, c) mean of 7 dams

TABLE 3.5.3.3
PREGNANCY STATUS IN RATS TREATED ORALLY WITH OLMESARTAN MEDOXOMIL AND C-SECTIONED ON DAY 20 OF GESTATION

Dose (mg/kg)	Number of pregnant females	Number of corpora lutea (Mean±S.E.M.)	Number of implantations (Mean±S.E.M.)	Number of living fetuses (Mean±S.E.M.)	Pre-implantation loss[1] (%)	Post-implantation loss[2] (%)	Body weight of fetuses (g) (Mean±S.E.M.)	Sex ratio (Male/Female)	Type and number of external malformations (%)
Control	25	444 (17.8 ± 0.42)	419 (16.8 ± 0.38)	399 (16.0 ± 0.44)	25(5.6)	20(4.8)	3.83 ± 0.034	0.99 (198/201)	0
40	25	463 (17.8 ± 0.39) P=0.9339	431 (16.6 ± 0.36) P=0.7279	415 (16.0 ± 0.43) P=0.9980	32(6.9) P=0.4269	16(3.7) P=0.4426	3.77 ± 0.044 P=0.2639	1.13 (220/195) P=0.3337	1 (0.2) a) P=0.9843
200	25	422 (16.9 ± 0.61) P=0.1902	396 (15.8 ± 0.53) P=0.1638	370 (14.8 ± 0.54) P=0.1034	26(6.2) P=0.7403	26(6.6) P=0.2678	3.71 ± 0.040 P=0.0197	0.97 (182/188) P=0.9041	0 ND
1000	20	357 (17.9 ± 0.42) P=0.8812	335 (16.8 ± 0.38) P=0.9846	310 (15.5 ± 0.51) P=0.4935	22(6.2) P=0.7502	25(7.5) P=0.1214	3.46 ± 0.066* P→0.0000	1.00 (155/155) P=0.9209	1 (0.3) b) P=0.8992

1) (number of corpora lutea – number of implantations) / number of corpora lutea X 100
2) (number of implantations – number of living fetuses) / number of implantations X 100
* Significant at 1% level compared to control. The decrease in body weight of fetuses at 200 mg/kg/day is significant at 5% level.
a) micro- or anophthalmia
b) omphalocele

TABLE 3.5.3.4
VISCERAL OBSERVATIONS IN FETUSES DERIVED FROM RAT DAMS TREATED ORALLY WITH OLMESARTAN MEDOXOMIL

Dose (mg/kg)	Number of fetuses examined	Type and number of malformations (%)	Type and number of variations (%)		
			Thymic remnant in neck	Dilatation of renal pelvis	Left umbilical artery
Control	194	3(1.5) a)3	22(11.3)	12(6.2)	5(2.6)
40	199	8(4.0) a)3 b)1 c)1 d)3 P=0.2378	11(5.5) P=0.0378	12(6.0) P=0.9487	4(2.0) P=0.9692
200	178	6(3.4) a)4 d)2 P=0.4201	14(7.9) P=0.2574	22(12.4) P=0.0390	0 P=0.0881
1000	150	3(2.0) a)2 e)1 P=0.9231	7(4.7) P=0.0272	27(18.0) P=0.0304	0 P=0.1269

a) ventricular septal defect
b) abnormal origin of right subclavian artery
c) right subclavian artery arising from aortic arch
d) dilatation of lateral ventricle
e) atrial septal defect

In the macroscopic examination of the thoracic/peritoneal viscera of F_0 dams, 2 of the high dose group dams showed petechia or ecchymoses (2 mm size) in the region of the gastric fundus gland. No abnormalities were observed in any other treatment group.

Among dams allocated to natural delivery, the period of pregnancy and the number of implantation scars in the treatment groups did not differ significantly from the control group. No abnormalities in external appearance were observed. One high dose dam delivered on day 21 of pregnancy. Since all of her pups were stillborn, the dam was autopsied. A significant treatment-related decrease ($p < 0.05$) in mean birth weight was observed in offspring that had been exposed, in utero, to 200 or more mg/kg/day (Table 3.5.3.5). A statistically significant ($p < 0.05$) decrease in pup survival rate to day 4 was observed in all dose groups relative to the control group. Survival of mid dose pups to weaning was decreased (Table 3.5.3.6) but the difference from control was not significant ($p > 0.05$) and was attributed to one dam that was sacrificed in poor condition on lactation day 11 (all of her pups sacrificed). In comparison with the control group, significant ($p < 0.05$) decreases in F_1 body weight were observed between pp days 22 and 57 in the 200 and 1000 mg/kg/day groups (both males and females) and between pp days 43 and 57 days in the 40 mg/kg/day group (males only) (Tables 3.5.3.7 and 3.5.3.8). Body weights on day

57 were 94.1% and 94.8% of control for males and females, respectively, in the 200 mg/kg/day group and were 90.7% and 94.2% of control for males and females, respectively in the 1000 mg/kg/day group (Table 3.5.3.8).

TABLE 3.5.3.5
REPRODUCTIVE PERFORMANCE OF F_0 FEMALES THAT DELIVERED NATURALLY AND SUMMARY OF STATUS OF F_1 GENERATION AT BIRTH

Dose (mg/kg)	Number of pregnant females	Number of delivered females (%)	Duration of pregnancy (days)	Number of implantations (Mean±S.E.M.)	Number of newborns (Mean±S.E.M.)	Parturition rate[1] (%)	Sex ratio (Male/Female)	Body weight of newborns (g) (Mean±S.E.M.)	Type and number of external malformations
Control	13	13 (100.0)	21.6	199 (15.3 ± 0.72)	186 (14.3 ± 0.65)	93.5	0.96 (91 / 95)	7.3 ± 0.24	0/ 186
40	13	13 (100.0) ND	21.6 ND	205 (15.8 ± 0.94) P=0.7004	183 (14.1 ± 0.82) P=0.8278	89.3 P=0.1336	1.03 (93 / 90) P=0.7159	7.0 ± 0.30 P=0.4287	0/ 183 ND
200	13	13 (100.0) ND	21.6 P=0.4517	218 (16.8 ± 0.44) P=0.0961	183 (14.1 ± 0.67) P=0.8081	83.9 P=0.2219	1.20 (100 / 83) P=0.2716	6.2 ± 0.35 P=0.0197	0/ 183 ND
1000	13	9 (69.2) P=0.1030	21.6 P=0.7914	135 (15.0 ± 0.71) P=0.7720	113 (12.6 ± 1.66) P=0.3470	83.7 P=0.4088	1.31 (64 / 49) P=0.1956	6.0 ± 0.54 P=0.0209	0/ 113 ND

1) (NUMBER OF NEWBORNS / NUMBER OF IMPLANTATIONS) X 100

TABLE 3.5.3.6
SURVIVAL RATES OF F_1 GENERATION DERIVED FROM DAMS TREATED WITH OLMESARTAN MEDOXOMIL

Dose (mg/kg)	Number of newborns (Mean per dam)	Number of neonates at 4th day (%)	Number of dead neonates up to 4th day (%)	Number of nurslings after selection at 4th day (Mean±S.E.M.)	Number of weanlings (%)
Control	186(14.3)	186(100.0)	0(0.0)	104(8.0±0)	104(100.0)
40	183(14.1) P=0.8278	176(96.2) P=0.0208	7(3.8) ND	102(7.8±0.15) P=0.3370	102(100.0) ND
200	183(14.1) P=0.8081	172(94.0) P=0.0603	11(6.0) ND	96(8.0±0) ND	88(91.7) P=0.2980
1000	113(12.6) P=0.3470	94(83.2) P=0.0480	19(16.8) ND	56(8.0±0) ND	56(100.0) ND

TABLE 3.5.3.7

BODY WEIGHT CHANGES IN F_1 GENERATION DERIVED FROM DAMS TREATED WITH OLMESARTAN MEDOXOMIL. POSTNATAL DAYS 4-22.

Dose (mg/kg)	Sex	Number of dams	Number of nurslings	Number of weanlings	Body weight (g) at each postnatal day			
					4	8	15	22
Control	Male	13	53	53	10.7 ± 0.47	20.4 ± 0.58	36.9 ± 0.67	65.3 ± 1.07
	Female	13	50	50	10.2 ± 0.39	19.7 ± 0.44	37.5 ± 0.57	62.0 ± 1.03
40	Male	13	52	52	ND 10.7 ± 0.52	P=0.9870 20.4 ± 0.73	P=0.6490 39.4 ± 0.96	P=0.7520 64.6 ± 1.85
	Female	13	50	50	P=0.8997 10.1 ± 0.46	P=0.8004 19.5 ± 0.65	P=0.7998 37.7 ± 0.88	P=0.9908 61.9 ± 1.68
200	Male	12	49	45 a)	P=0.1520 9.6 ± 0.56	P=0.1674 18.3 ± 1.30	P=0.3128 37.7 ± 0.88	P=0.0285 61.6 ± 1.19
	Female	12	47	43 b)	P=0.1024 9.1 ± 0.58	P=0.0981 17.3 ± 1.30	P=0.1480 35.8 ± 0.99	P=0.0351 58.0 ± 1.52
1000	Male	7	30	30	P=0.1029 9.1 ± 0.90	P=0.1680 17.7 ± 1.64	P=0.1526 35.7 ± 1.87	P=0.0558 58.5 ± 2.86
	Female	7	26	26	P=0.1424 9.0 ± 0.85	P=0.1939 17.5 ± 1.46	P=0.1518 35.5 ± 1.45	P=0.0399 57.4 ± 2.12

Data are expressed as mean ± SEM

a) Four pups died on 6, 8, 10 and 11 days after birth

b) Four pups died on 6, 9, 10 and 11 days after birth

TABLE 3.5.3.8

BODY WEIGHT CHANGES IN F_1 GENERATION DERIVED FROM DAMS TREATED WITH OLMESARTAN MEDOXOMIL. POSTNATAL DAYS 29-57.

Dose (mg/kg)	Sex	Number of animals	Body weight (g) at each postnatal day				
			29	36	43	50	57
Control	Male	53	116.0 ± 1.05	181.9 ± 1.63	248.5 ± 1.91	309.3 ± 2.74	369.0 ± 2.59
	Female	50	104.8 ± 1.11	149.9 ± 1.66	186.1 ± 1.89	212.6 ± 2.97	241.4 ± 2.70
40	Male	52	P=0.1494 113.2 ± 1.63	P=0.0948 177.1 ± 2.33	P=0.0117 239.8 ± 2.76	P=0.0218 299.3 ± 3.35	P=0.0184 357.8 ± 3.89
	Female	50	P=0.6823 104.0 ± 1.41	P=0.8548 150.3 ± 1.78	P=0.8442 185.6 ± 1.97	P=0.8981 213.1 ± 2.44	P=0.8484 240.7 ± 2.47
200	Male	45	* P=0.0001 109.0 ± 1.25	* P=0.0002 172.1 ± 1.70	* P→0.0000 234.1 ± 2.19	* P=0.0003 294.3 ± 2.75	* P→0.0000 347.3 ± 3.30
	Female	43	* P→0.0000 96.9 ± 1.22	* P=0.0001 141.7 ± 1.48	* P=0.0002 176.6 ± 1.94	P=0.0226 203.5 ± 2.41	* P=0.0004 228.6 ± 2.77
1000	Male	30	* P→0.0000 100.6 ± 2.68	* P→0.0000 162.4 ± 3.64	* P→0.0000 223.5 ± 4.42	* P=0.0001 282.5 ± 5.50	* P→0.0000 334.7 ± 5.62
	Female	26	* P=0.0001 95.7 ± 1.93	* P=0.0006 140.8 ± 1.87	* P→0.0000 173.5 ± 2.25	* P=0.0019 200.5 ± 2.71	* P=0.0004 227.2 ± 2.56

Data are expressed as mean ± SEM

* Significant at 1% level compared with control

In the observation of postnatal differentiation, which is one of the indices of postnatal growth, separation of the ear auricula, eruption of lower incisors, appearance of abdominal hair, and separation of eyelids tended to be delayed in pups from the 200 and 1000 mg/kg/day groups, but the differences from the control group were not statistically significant ($p > 0.05$). Opening of the vagina tended to be delayed in pups from the 1000 mg/kg/day group. No treatment-related effects were observed for results of the righting reflex test, negative geotaxis test, free-fall righting reflex test, sensory function test, emotionality test, or learning test.

All mated female F_1 rats from the treated groups became pregnant and there were no differences in the length of the gestation period or mean pup body weights at birth. A statistically significant ($p < 0.05$) decrease in the number of implantation scars was noted for F_1 dams from the 1000 mg/kg/day group. Also, numbers of F_2 pups delivered by dams from the 200 and 1000 mg/kg/day groups were significantly ($p < 0.05$) reduced (Table 3.5.3.9). There was no effect of treatment on F_2 body weights or external appearance. No macroscopic abnormalities were observed at necropsy of the F_1 animals and there were no abnormalities of sperm or sperm motility.

TABLE 3.5.3.9
REPRODUCTIVE PERFORMANCE OF F_1 FEMALES AND SUMMARY OF STATUS OF F_2 GENERATION PRIOR TO WEANING

Dose (mg/kg)	Number of pairs	Number of copulated females	Number of pregnant females	Number of delivered females (%)	Duration of pregnancy (days)	Number of implantations (Mean±S.E.M.)	Number of newborns (Mean±S.E.M.)	Sex ratio (Male/Female)	Body weight of newborns (g) (Mean±S.E.M.)	Type and number of external malformations
Control	10	10	6	6 (100.0)	21.8	110 (18.3 ± 0.56)	105 (17.5 ± 0.67)	1.23 (58 / 47)	6.9 ± 0.16	0/ 105
40	10	10	10	10 (100.0) P=0.0935	22.0 P=0.3632	173 (17.3 ± 0.80) P=0.3773	161 (16.1 ± 1.03) P=0.3464	0.94 (78 / 83) P=0.2788	6.8 ± 0.23 P=0.8881	0/ 161 ND
200	10	10	10	10 (100.0) P=0.0935	21.7 P=0.5816	170 (17.0 ± 0.60) P=0.1551	152 (15.2 ± 0.47) P=0.0117	0.88 (71 / 81) P=0.1789	6.8 ± 0.17 P=0.7643	0/ 152 ND
1000	10	10	10	10 (100.0) P=0.0935	21.8 P=0.8792	155 (15.5 ± 0.73) P=0.0173	139 (13.9 ± 0.81)* P=0.0043	1.07 (72 / 67) P=0.5939	7.5 ± 0.37 P=0.1762	0/ 139 ND

* Significant at 1% level compared with control

Based on the results of this study, 200 mg/kg/day was considered to be a toxic dose for dams and for fetal growth. The no-effect dose for general toxicity, development, and reproduction and development of the F_1 generation is considered to be 40 mg/kg/day.

3.5.4. Developmental Toxicity (Segment II) Study in Rabbits. (Study #95-0052, Report #TR 142-147). Vol. 26

This GLP study was conducted by the Laboratory Animal Science and Toxicology Laboratories, Sankyo Co., Ltd., Shizuoka, Japan, between July 20, 1995 and July 22, 1996. Dosing and necropsy dates are not given. The study investigated the effects of olmesartan medoxomil (OM) during organogenesis.

Animals

Japanese White rabbits were 6 to 7 months of age and weighed between 2.73 and 3.59 kg on day of copulation. Animals were housed individually in metallic cages.

Mode of Administration/Dosage Levels

Suspensions of OM (lot #NH006C1) were prepared in 0.5% CMC and administered orally, by gavage (3 ml/kg), once daily, to groups of mated females at doses of 0.3, 1 or 3 mg/kg (17, 25 and 9 rabbits, respectively, became pregnant) on gestation days 6 to 18. Control animals (13 of which became pregnant), received the vehicle.

The doses were selected based on a preliminary study in which olmesartan was administered at doses of 1, 3 or 10 mg/kg to pregnant rabbits (5/group) from days 6 to 18 of pregnancy. One of 5 and three of 5 rabbits in the mid and high dose groups, respectively, died after showing remarkable decreases in food intake and body weight gain. At the low dose, a slight decrease in food intake was observed in 3 of 5 rabbits; however, there were no adverse effects on general condition and body weight gain. No adverse effects on fetuses were noted in any group.

Observations/Measurements

All animals were observed daily. Body weights and food intake were recorded days 0 to 28 of pregnancy. On day 28 of pregnancy, surviving dams were euthanized under anesthesia (pentobarbital sodium). The organs in the thoracic and peritoneal cavities were examined macroscopically. Thereafter, the numbers of corpus lutea, implantations, dead embryos/fetuses, and live fetuses were recorded. The live fetuses were sexed, weighed, and macroscopically examined for external abnormalities. Fetuses with a normal external appearance were autopsied and organs in the thoracic and peritoneal cavities were examined macroscopically. These fetuses were then processed into skeletal specimens and examined for skeletal abnormalities. Except for two fetuses in the mid dose group, in which skeletal examinations were performed, all fetuses with abnormal external appearance were stored as specimens and not further examined.

Results

Eight of the nine pregnant does in the high dose group died between gestation days 12 and 17 (2 on day 12, 3 each on day 16 and 17). All of them showed a remarkable decrease in food intake (stopped eating 1-5 days before death), a slight decrease in body weight gain, soft feces, a decrease in locomotor activity and a staggering gait. Autopsy revealed stomach ulcer in all of the

deceased animals. The lone survivor showed no abnormality in its general condition. Thus, the evaluation of effects on pregnancy was limited to low dose and mid dose groups. In the mid dose group (1 mg/kg/day), 7 does died between day 14 and day 20 of pregnancy (1 each on day 14 and 19, 2 on day 20, 3 on day 17). As in high dose group animals, stomach ulcer was observed in all dead dams. Daily food intake (but not body weight gain) was statistically significantly lower ($p < 0.05$) than control from day 11 to day 16 of pregnancy (Table 3.5.4.1). A slight decrease in food intake was also observed for the low dose group but it did not attain a level of statistical significance ($p > 0.05$). No animal died in the control and low dose groups.

TABLE 3.5.4.1
FOOD INTAKE IN RABBIT DAMS TREATED ORALLY WITH OLMESARTAN MEDOXOMIL

Dose	Food intake (g) on each gestational day										
mg/kg	1	6	11	12	13	14	15	16	17	18	19
0	133.6	140.5	139.8	137.5	133.5	134.5	131.2	135.5	139.0	140.8	138.6
0.3	129.1	143.9	136.9	132.1	122.9	122.0	123.8	126.4	128.9	131.3	125.5
1	135.5	146.7	122.0*	116.0*	109.6*	108.7*	103.7*	100.3*	117.1	117.3	116.7

Data expressed as mean. *: Significantly different from control ($p < 0.05$).

Numbers of corpora lutea, implantations and living fetuses were similar in drug treated and control groups. However, preimplantation loss in the 1 mg/kg/day group (10.9%) was significantly ($p < 0.05$) higher than in the control group (3.4%). Significantly ($p < 0.05$) increased fetus weights were observed in this group relative to the control group (Table 3.5.4.2).

TABLE 3.5.4.2
PREGNANCY STATUS IN RABBIT DAMS TREATED ORALLY WITH OLMESARTAN MEDOXOMIL

Dose (mg/kg)	Number of dams at term	Number of corpora lutea (Mean±S.E.M.)	Number of implantations (Mean±S.E.M.)	Number of living fetuses (Mean±S.E.M.)	Pre-implantation loss[1] (%)	Post-implantation loss[2] (%)	Body weight of fetuses (g) (Mean±S.E.M.)	Sex ratio (Male/Female)
0	13	117 (9.0 ± 0.32)	113 (8.7 ± 0.26)	98 (7.5 ± 0.46)	4 (3.4)	15 (13.3)	35.9 ± 0.78	0.78 (43 / 55)
0.3	15	133 (8.9 ± 0.51) P=0.8316	128 (8.5 ± 0.52) P=0.7861	114 (7.6 ± 0.52) P=0.9314	5 (3.8) P=0.8446	14 (10.9) P=0.5779	37.3 ± 1.13 P=0.3399	1.24 (63 / 51) P=0.0983
1	17	147 (8.6 ± 0.36) P=0.4874	131 (7.7 ± 0.59) P=0.1390	117 (6.9 ± 0.58) P=0.4065	16 (10.9) P=0.0410	14 (10.7) P=0.5335	40.5 ± 0.97 P=0.0146	1.13 (62 / 55) P=0.1830

1) $\frac{\text{(number of corpora lutea – number of implantations)}}{\text{number of corpora lutea}} \times 100$

2) $\frac{\text{(number of implantations – number of living fetuses)}}{\text{number of implantations}} \times 100$

No significant differences from control in the incidence of external, visceral or skeletal anomalies were observed in fetuses in the 0.3 and 1 mg/kg/day groups. The abnormal external appearances observed in different litters are given in the Table 3.5.4.3. Skeletal anomalies are listed in the Table 3.5.4.4. There were no visceral anomalies reported in any group. In conclusion, olmesartan medoxomil exhibited no evidence of embryo-fetal toxicity in the

pregnant rabbit. The no adverse effect dose of OM for the rabbit doe and for development of the next generation (in this study) was 0.3 mg/kg.

TABLE 3.5.4.3
EXTERNAL OBSERVATIONS IN FETUSES DERIVED FROM EACH RABBIT TREATED ORALLY WITH OLMESARTAN MEDOXOMIL

Animal No.	Number of fetuses examined	External malformations total	Acephaly and thoraco-gastroschisis	Cleft palate	Encephalocele	Thoraco-gastroschisis	Brachyury	Nasal hypoplasia and omphalocele	Club foot	Nasal hypoplasia and spina bifida
0 mg/kg										
1	4	1 (25.0)	1 (25.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
2	10	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
3	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
4	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
5	8	1 (12.5)	0 (0.0)	1 (12.5)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
6	7	1 (14.3)	0 (0.0)	0 (0.0)	1 (14.3)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
7	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
8	9	1 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)	1 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
102	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
103	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
104	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
105	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
106	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	98	4 (4.8)	1 (1.9)	1 (1.0)	1 (1.1)	1 (0.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
0.3 mg/kg										
21	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
23	11	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
24	5	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
25	Exclueded due to splitting of uterine horn									
26	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
27	12	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
28	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
201	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
202	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
204	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
205	5	1 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)
206	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
208	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
209	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
210	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
211	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	114	1 (1.3)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.3)	0 (0.0)	0 (0.0)	0 (0.0)
1 mg/kg										
41	9	1 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (11.1)	0 (0.0)	0 (0.0)
42	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
44	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
46	4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
402	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
403	12	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
404	5	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
405	Exclueded due to splitting of uterine horn									
408	2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
409	5	1 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (20.0)	0 (0.0)
410	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
411	4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
412	9	1 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (11.1)
415	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
418	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
419	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
421	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
423	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	117	3 (2.5)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.7)	1 (1.2)	1 (0.7)

TABLE 3.5.4.4

SKELETAL OBSERVATIONS IN FETUSES DERIVED FROM EACH RABBIT TREATED ORALLY WITH OLMESARTAN MEDOXOMIL

Animal No.	Number of fetuses examined	Malformations: Total	Malformations: Fusion and absence of thoracic vertebral arches and fusion of ribs	Malformations: Absence of ribs	Variations: 13th rib	Variations: Irregular alignment of caudal vertebrae	Variations: 27 presacral vertebrae	Variations: Asymmetry of sternebrae
0 mg/kg								
1	3	0 (0.0)	0 (0.0)	0 (0.0)	1 (33.3)	0 (0.0)	1 (33.3)	0 (0.0)
2	10	0 (0.0)	0 (0.0)	0 (0.0)	2 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)
3	7	0 (0.0)	0 (0.0)	0 (0.0)	1 (14.3)	0 (0.0)	0 (0.0)	0 (0.0)
4	6	0 (0.0)	0 (0.0)	0 (0.0)	1 (16.7)	1 (16.7)	0 (0.0)	0 (0.0)
5	7	0 (0.0)	0 (0.0)	0 (0.0)	3 (42.9)	0 (0.0)	0 (0.0)	0 (0.0)
6	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
7	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
8	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
102	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
103	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
104	7	0 (0.0)	0 (0.0)	0 (0.0)	3 (42.9)	1 (14.3)	0 (0.0)	1 (14.3)
105	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
106	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	94	0 (0.0)	0 (0.0)	0 (0.0)	11 (13.1)	2 (2.4)	1 (2.6)	1 (1.1)
0.3 mg/kg								
21	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
23	11	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
24	5	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
25	Excluded due to splitting of uterine horn							
26	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
27	12	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
28	8	0 (0.0)	0 (0.0)	0 (0.0)	1 (12.5)	0 (0.0)	0 (0.0)	0 (0.0)
201	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
202	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (12.5)	0 (0.0)
204	7	0 (0.0)	0 (0.0)	0 (0.0)	2 (28.6)	0 (0.0)	0 (0.0)	0 (0.0)
205	4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
206	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
208	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (12.5)	0 (0.0)	0 (0.0)
209	9	0 (0.0)	0 (0.0)	0 (0.0)	1 (11.1)	0 (0.0)	1 (11.1)	0 (0.0)
210	7	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
211	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	113	0 (0.0)	0 (0.0)	0 (0.0)	4 (3.5)	1 (0.8)	2 (1.6)	0 (0.0)
1 mg/kg								
41	8	3 (37.5)	1 (12.5)	2 (25.0)	3 (37.5)	0 (0.0)	5 (62.5)	0 (0.0)
42	8	0 (0.0)	0 (0.0)	0 (0.0)	1 (12.5)	0 (0.0)	0 (0.0)	0 (0.0)
44	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
46	4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
402	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
403	12	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
404	5	0 (0.0)	0 (0.0)	0 (0.0)	1 (20.0)	0 (0.0)	1 (20.0)	0 (0.0)
405	Exoluded due to splitting of uterine horn							
408	2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
409	4	0 (0.0)	0 (0.0)	0 (0.0)	1 (25.0)	0 (0.0)	0 (0.0)	0 (0.0)
410	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
411	4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
412	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
415	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
418	8	0 (0.0)	0 (0.0)	0 (0.0)	2 (25.0)	0 (0.0)	1 (12.5)	0 (0.0)
419	8	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
421	7	0 (0.0)	0 (0.0)	0 (0.0)	2 (28.6)	0 (0.0)	1 (14.3)	0 (0.0)
423	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (16.7)	0 (0.0)	0 (0.0)
	114	3 (2.2)	1 (0.7)	2 (1.5)	10 (8.7)	1 (1.0)	8 (6.4)	0 (0.0)

3.5.5. Late Gestation and Lactation (Segment III) Study in Rats (Study #95-0040, Report #TR 143-010). Vol. 26.

This GLP study, conducted by the Laboratory Animal Science and Toxicology Laboratories, Sankyo Co., Ltd., Shizuoka, Japan, investigated the effects of olmesartan medoxomil on fetal development, parturition, lactation, neonatal growth, development and survival in rats. Dosing was completed on July 22, 1995.

Animals

Female Crj:CD rats [redacted] were approximately 10 weeks of age and weighed between 220 and 266 g on day 0 of gestation. Animals were housed individually with *ad libitum* access to food and water.

Mode of Administration/Dosage Levels

Suspensions of olmesartan medoxomil (lot #NH006C1) were prepared in 0.5% CMC and administered orally by stomach tube (0.5 ml/100 g), once daily, to groups of mated females at doses of 8, 40 or 200 mg/kg (22, 22 and 24 rats, respectively, became pregnant) from gestation day 17 to lactation day 21. Control animals (25 of which became pregnant) received the vehicle.

Dose selection was based on a developmental toxicity study (report #TR 141-086, section #3.5.3) and a preliminary study of perinatal and lactational toxicity in rats (report #TR 142-068). In both studies, reduced maternal food intake and body weight gain were observed at doses 200 or more mg/kg/day. In the first study, 4 of 13 dams receiving 1000 mg OM/kg/day died (2 on day 22 of gestation, one during labor on day 21 of pregnancy and one on day 11 postpartum) as did one of 13 dams receiving 200 mg/kg/day. In the second study, one of 9 dams each at 200 and 1000 mg/kg/day died during parturition and on day 22 of pregnancy, respectively.

Observations/Measurements: All animals were observed daily. Body weights were recorded on gestation days 0, 7, 9, 11, 13, 15, 17-20, and lactation days 1, 4, 8, 15 and 22. Food consumption was determined on days of body weight measurement except on day 0 of pregnancy and day 1 of parturition. All dams were allowed to undergo natural delivery. If rats showed no signs of delivery by day 25 of gestation, they were autopsied and the uterus was examined for evidence of pregnancy. Duration of gestation, litter size, stillbirths and live births, sex and the presence of any external abnormal appearance were recorded.

On lactation day 4, each litter was reduced to 4 males and 4 females. The offspring were weaned on day 22 and the weaning index (ratio of the number of live newborns on day 22 to the number of live pups on day 4) was calculated. The dams were autopsied at weaning. After macroscopic examination of thoracic/peritoneal viscera, the implantation scars in the uteri of the dams were counted and the birth index (ratio of the number of live newborns to the number of implantation scars) was calculated. Newborns were weighed on postpartum days 1, 4, 8, 15, 22, 29, 36, 43, 50 and 57. During this period, F_1 pups were observed for maturation (lower incisor eruption, separation of ear auricula, appearance of abdominal hair, separation of eyelids, testes descent, vaginal opening). Further, the animals were tested for the sensory function (pupillary, corneal

and Preyer's reflexes), emotional (open field performance) and learning (conditioned avoidance response) ability, behavior (maze and motility) and exercise performance (righting reflex, negative geotaxis and free-fall righting reflex).

At 10 weeks of age, F_1 animals (10 per sex per group were selected at 8 weeks) were cohabited (1 male and 1 female per litter but avoiding sib mating) within the same treatment group for 14 days to examine their reproductive capability. Each dam was allowed to undergo natural parturition, and the birth index and the gestation period were determined. One day after parturition, F_2 pups were counted, weighed, sexed and examined for external abnormalities. F_1 dams were sacrificed after delivery to examine thoracic/peritoneal viscera macroscopically and implantation scars in the uteri were counted. In the case of males, the absence or presence of sperm in the epididymis was determined and their motility examined. The F_1 animals not used for the fertility examination were killed at 8 weeks of age, and thoracic/peritoneal viscera macroscopically examined.

Results

There were no mortalities. There were no test substance-related physical signs except for piloerection observed in 2 and 4 dams in the 40 and 200 mg/kg/day dose groups, respectively, from 4 to 9 days after parturition. No significant reduction in body weight gain was observed during the pregnancy period in any treated group relative to the control group. However, between lactation days 4 and 8, a significant reduction in body weight gain was observed for dams receiving 200 mg/kg/day (Table 3.5.5.1). Food consumption was lower than control in the mid and high dose groups. The difference was significant on day 20 of pregnancy in the 40 (-12%) and 200 (-13%) mg/kg/day treated groups, and 4, 8, 15 and 22 days (-31, -25, -13 and -24%, respectively) after parturition in the 200 mg/kg/day treated groups (Table 3.5.5.2).

TABLE 3.5.5.1
PERINATAL & LACTATIONAL PERIODS IN RATS: MEAN BODY WEIGHT GAINS OF F_0 DAMS

Dose, mg/kg	# pregnant females	B.wt on GD 0	Body weight gain (% increase relative to GD 0)						
			GD 18	GD 20	LD 1	LD 4	LD 8	LD 15	LD 22
0	24	238.1	52.0	66.0	20.0	30.7	35.5	38.4	28.0
8	22	238.6	54.0	69.0	21.5	30.0	35.3	40.2	29.5
40	22	243.8	52.4	63.5	18.5	24.6	33.0	38.8	28.9
200	24	237.3	54.2	65.3	18.6	20.9*	26.0*	37.5	27.5

* $p < 0.01$ compared with control

All pregnant dams delivered after a normal gestation period (21.3 to 21.6 days). No abortions or resorptions were noted; litter parameters were comparable in the control and treated groups (Table 3.5.5.3). There were no treatment-related external abnormalities observed in the newborns. Macroscopic examination of dams sacrificed during weaning revealed dark-red hemorrhagic spots at the stomach fundus glandularis in one high dose group animal. No abnormalities were observed in any other group.

TABLE 3.5.5.2
PERINATAL & LACTATIONAL PERIODS IN RATS: MEAN FOOD INTAKE OF F_0 DAMS

Dose (mg/kg)	Number of pregnant females	Food intake (g) on each gestational day						Number of nursing dams	Food intake (g) on each day after delivery			
		7	14	17	18	19	20		4	8	15	22
0	24	27.0 ±0.53	26.3 ±0.61	28.8 ±0.43	20.3 ±0.76	23.5 ±0.67	22.2 ±0.45	24	40.5 ±1.65	48.6 ±1.46	59.4 ±1.22	75.0 ±1.46
8	22	27.0 ±0.53 ND	27.1 ±0.58 P=0.3482	28.4 ±0.62 P=0.6108	22.5 ±0.86 P=0.0554	23.4 ±0.56 P=0.9558	22.2 ±0.58 P=0.9342	21	40.1 ±1.19 P=0.8802	46.1 ±0.90 P=0.1564	60.0 ±1.12 P=0.7088	74.8 ±1.34 P=0.9085
40	22	28.1 ±0.55 P=0.1611	27.6 ±0.68 P=0.1595	29.8 ±0.65 P=0.2066	20.9 ±0.66 P=0.5771	22.7 ±0.62 P=0.4305	19.5* ±0.59 P=0.0013	22	34.4 ±2.20 P=0.0299	43.9 ±1.14 P=0.0157	55.5 ±1.16 P=0.0269	71.8 ±2.47 P=0.2629
200	24	27.1 ±0.48 P=0.9083	26.7 ±0.49 P=0.6332	28.5 ±0.46 P=0.6000	22.0 ±0.93 P=0.1729	23.0 ±0.69 P=0.6664	19.3* ±0.77 P=0.0055	24	28.0* ±2.68 P=0.0005	36.3*a) ±2.54 P→0.0000	51.5*b) ±1.69 P=0.0005	57.1* ±2.33 P→0.0000

Data expressed as mean ± S.E.M.
*: Significant at 1% level compared with control
a) mean of 23 dams, b) mean of 22 dams

TABLE 3.5.5.3
PERINATAL & LACTATIONAL PERIODS IN RATS: SURVIVAL RATES OF OFFSPRING

Parameters	Control	8 mg/kg/day	40 mg/kg/day	200 mg/kg/day
No. of delivered females (%)	24 (100)	22 (100)	22 (100)	24 (100)
No. of implantations (mean)	373 (15.5)	360 (16.4)	364 (16.5)	380 (15.8)
No. of newborns (mean)	332 (13.8)	337 (15.3)	325 (14.8)	357 (14.9)
Parturition rate, %	89	93.6	89.3	93.9
No. of pups on day 4 (%)	319 (96.1)	318 (94.4)	303 (93.2)	328 (91.9)
No. of pups after redn. (mean)	187 (7.8)	168 (8.0)	175 (8.0)	178 (7.4)
No. of weanlings (%)	185 (98.9)	168 (100)	171 (97.7)[a]	160 (89.9)[b]

a: 4 pups died 6, 9, 10 and 11 days after birth
b: 18 pups died 6, 7, 8, 10, 21 and 22 days after birth

F_1 generation: Piloerection, decreased spontaneous locomotor activity, staggering gait and tremor were observed around the weaning period (21 to 23 days after birth) in 2 and 6 pups in the mid and high dose groups, respectively; all of these pups died before lactation day 24. An additional five pups from the 200 mg/kg/day group died just before weaning, although no clinical signs were apparent. Macroscopic examination at the unscheduled autopsies of F_1 pups revealed dark-red hemorrhagic spots at the stomach fundus glandularis in 1 mid dose and 1 high dose pup, and dilatation of the renal pelvis in 1 mid dose and 5 high dose pups. Because of these deaths, the survival rate of F_1 pups to day 4 (91.9%) and from lactation day 4 to lactation day 22 (89.9%) in the high dose group was lower than control (96.1% and 98.9%, respectively) but the difference was not statistically significant (Table 3.5.5.3). Statistically significant treatment-

related reductions in birth weight in all dose groups were observed, as were reductions in weight gain of pups throughout lactation and post weaning thorough day 50 postpartum (Table 3.5.5.4). Mean body weights of the newborn were 92.5%, 91%, and 88% of control value in the 8, 40, and 200 mg/kg/day groups, respectively. The body weights were generally comparable to control values by pp day 57.

TABLE 3.5.5.4
PERINATAL & LACTATIONAL PERIODS IN RATS: MEAN F_1 BODY WEIGHT (GM)

Dose, mg/kg	B.wt at birth[a]	Postnatal Day								
		4	8	15	22	29	36	43	50	57
0	6.7	M 9.9	19.2	37.2	60.6	104.7	163.7	225.6	283.6	335.3
		F 9.5	18.6	36.3	59.0	96.4	139.0	174.3	200.1	220.9
8	6.2*	M 9.0*	17.9§	35.3*	56.9*	99.7*	157.7*	217.5*	274.8*	327.4*
		F 8.7§	17.4§	34.2*	54.8*	91.9*	133.7*	167.9*	193.8*	216.6
40	6.1§	M 9.0	17.0§	33.7*	54.7*	101.1§	161.5	224.1	284.2	337.0
		F 8.7	16.5§	32.7*	52.8*	91.9*	135.3§	170.3§	197.2	220.9
200	5.9*	M 8.6*	16.0*	31.4*	45.2*	90.0*	149.4*	213.9*	275.2§	331.4
		F 8.1*	15.3*	30.0*	44.3*	82.4*	127.0*	164.8*	192.3*	217.3

a: both sexes combined
* $p < 0.01$ compared with control; § $p < 0.05$ compared with control

Most developmental landmarks (separation of ear auricula, eruption of lower incisors, appearance of abdominal hair, separation of eyelids, descent of testes and opening of vagina) showed low frequencies of appearance in all drug treated groups at the initial observation. However, at the second observation (made within 1 to 3 days), there were no significant differences from control. Prolongation of righting reflex latency ($p < 0.01$) was observed in females at 4 days of age in the 40 and 200 mg/kg/day groups (12.8 and 12.62 sec *vs.* 4.85 sec in control, respectively). The second motor coordination test, negative geotaxis, showed no positive reactions in mid and high dose males and females at 6 days of age, while 15% of the control animals responded. A low ratio of positive reactions ($p < 0.01$) was observed at 8 days of age in the 40 mg/kg/day (male, 30%, female, 5.3%) and 200 mg/kg/day (male, 0% and female, 5%) treated groups. Additionally, a significant ($p < 0.01$) difference in the reaction latency was also observed in both high and mid dose females (26 sec *vs* 16 sec for control). The test continued until 14 days of age when it was 100% positive in all treated groups. The free-fall righting reflex was negative for all high dose males and females at 14 days of age, while only 5 to 10% of low dose males and females responded (control 15 to 20%). At 18 days of age there was a positive reaction in all control and treated animals. The results of the sensory function tests showed no meaningful significant difference between control and treated groups. Results of learning, emotional and behavioral tests were similar in all groups.

OM had no adverse effects on the reproductive capacity of the F_1 animals. All pregnant dams showed normal parturition and no significant difference from control in number or weight of newborn. No abnormalities in external appearance were noted in the F_2 pups (Table 3.5.5.5).

TABLE 3.5.5.5
REPRODUCTIVE PERFORMANCE OF F_1 FEMALES AND SUMMARY STATUS OF F_2 GENERATION

Dose (mg/kg)	Number of pairs	Number of copulated females (%)	Number of pregnant females (%)	Number of delivered females (%)	Duration of pregnancy (days)	Number of implantations (Mean±S.E.M.)	Number of newborns (Mean±S.E.M.)	Sex ratio (Male/Female)	Body weight of newborns (g) (Mean±S.E.M.)	Type and number of external malformations
0	10	10(100.0)	10(100.0)	10 (100.0)	21.9	173 (17.3 ± 0.52)	159 (15.9 ± 0.50)	1.04 (81 / 78)	6.5 ± 0.19	0
8	10	9(90.0) P=1.0000	9(100.0) ND	9 (100.0) ND	21.8 P=0.4935	163 (18.1 ± 0.39) P=0.2353	149 (16.6 ± 0.47) P=0.3601	1.16 (80 / 69) P=0.6295	6.3 ± 0.26 P=0.5200	0 ND
40	10	8(80.0) P=0.4561	8(100.0) ND	8 (100.0) ND	21.6 P=0.1829	141 (17.6 ± 0.78) P=0.7236	132 (16.5 ± 0.80) P=0.5191	0.86 (61 / 71) P=0.4215	6.2 ± 0.21 P=0.3666	0 ND
200	10	9(90.0) P=1.0000	9(100.0) ND	9 (100.0) ND	21.7 P=0.2356	145 (16.1 ± 0.39) P=0.0890	131 (14.6 ± 0.41) P=0.0577	1.18 (71 / 60) P=0.5807	6.9 ± 0.20 P=0.1920	0 ND

Autopsy of F_1 adult animals revealed dilatation of the renal pelvis in 5 of 10 males and 1 of 10 females in the high dose group and 1 of 10 females in the mid dose group (Table 3.5.5.6). In addition, one high dose female showed white spots on the kidney. Histopathological examination of the white spots showed dilatation and cysts of the renal tubules, regeneration of the renal tubules and thickening of the basal membrane, necrosis of the renal tubular epithelium, interstitial cellular infiltration and fibrosis, confirming nephrosis. There was no abnormality in motility of epididymal sperm (examined in one male from each litter). F_1 animals not used for the fertility tests were killed at 8 weeks of age. In the males, dilatation of the renal pelvis was observed in 1/91, 1/73, 5/74, and 36/67 animals in the control, low, mid and high dose groups, respectively. In the females, dilatation of the renal pelvis was observed in 0/74, 8/75, 5/74, and 33/68 animals in the control, low, mid and high dose groups, respectively. Cystic kidney was observed in 1 male and 2 females in the 40 mg/kg/day group, and 1 male and 2 females in the 200 mg/kg/day group. Polycystic kidney was diagnosed in one high dose male. Additionally, 2 low dose females each showed a dark-red ulcer at the stomach fundus glandularis (Table 3.5.5.6).

TABLE 3.5.5.6
PERINATAL & LACTATIONAL PERIODS IN RATS: AUTOPSY OF F_1 ADULT ANIMALS (SCHEDULED SACRIFICE)

Observation	Control		8 mg/kg/day		40 mg/kg/day		200 mg/kg/day	
Sex	M	F	M	F	M	F	M	F
# examined	101	84	83	85	84	84	77	78
Dilatation of renal pelvis	1 (1)	0 (0)	1 (1.2)	8 (9.4)	5 (6)	6 (7.1)	41 (53.2)	34 (43.6)
Polycystic nephrosis	0 (0)	0 (0)	0 (0)	0 (0)	1 (1.2)	2 (2.4)	1 (1.3)	2 (2.6)
Ulcer[1]	0 (0)	0 (0)	0 (0)	2 (2.4)	0 (0)	0 (0)	0 (0)	0 (0)

[1]: ulcer with dark red hemorrhage at the stomach fundus glandularis.
Numbers in parentheses indicate percentage.

In summary, 200 mg OM/kg/day, administered from mid-gestation through lactation, significantly reduced maternal body weight gain and food intake during lactation. There were no abortions or resorptions. A lower F_1 birth weight in the high dose group (200 mg/kg/day) and body weight gain suppression during lactation and post weaning days in all treated groups were observed in comparison with the control group. The number of weanlings in the high dose group was significantly lower than control. A delay of postnatal maturation was noted in mid and high dose group pups. Fertility of the F_1 offspring was not impaired. The most notable finding was a significant increase in the rate of dilatation of the renal pelvis in the F_1 generation at all dose levels. A few F_1 animals in the mid and high dose groups displayed cystic kidney. The no effect dose level for abnormal development of the next generation was less than 8 mg/kg as the study failed to establish a no-effect level for effects on postnatal development.

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3.5.6. Late Gestation and Lactation (Segment III) Study in Rats. Supplementary Study with Lower Dosage Levels (Study # 96-0093, Report #TR-143-287). Vol. 27.

The Laboratory Animal Science and Toxicology Laboratories, Sankyo Co., Ltd., Shizuoka, Japan, conducted this GLP study between December 18, 1996 and March 18, 1998 (dates of dosing not provided). Since a no-effect level for effects on postnatal development was not observed in the previous study (section 3.5.5), this additional study was conducted in the same strain of rats using lower dosage levels.

Animals

Female Crj:CD rats [redacted] (28/dosage group) were approximately 10 weeks of age and weighed between 236 and 295 g on day of copulation. The animals were housed individually with *ad libitum* access to food and water.

Mode of Administration/Dosage Levels

Suspensions of olmesartan medoxomil (lot #NH006C1) were prepared in 0.5% CMC and administered orally *via* metallic stomach tube (0.5 ml/100 g), once daily, to mated females at doses of 0.3 or 1.6 mg/kg (26 and 28 of these rats, respectively, were pregnant) from gestation day 17 to lactation day 21. Control animals (27 of which were pregnant) received the vehicle.

The doses were selected based on the previous perinatal and lactational toxicity study in rats (TR 143-010). In that study there was retardation in postnatal development and an increased incidence of dilatation of the renal pelvis in the F_1 generation at all dose levels (doses as low as 8 mg/kg/day).

Observations/Measurements

As described in the previous section.

Results

There were no mortalities. No significant differences in body weight or food intake were observed in treated groups relative to the control group during the periods of pregnancy and lactation. All dams delivered after a normal gestation period. No abortions or resorptions were noted in any of the groups. Macroscopic examination of dams sacrificed at weaning revealed no abnormalities.

F_1 generation: No external abnormalities were observed in pups. The survival and weaning rates for the treated groups were not significantly different from those of the control group. According to the authors of the report, a significant decrease ($p < 0.01$) in body weight gain of the high dose group (relative to the control group) was observed post weaning only. However, reanalysis of the data by this reviewer showed a statistically significant decrease ($p < 0.05$) in body weight gain of the high dose group (relative to the control group) both pre- and post-weaning (Table 3.5.6.1). Regarding postnatal development, incomplete eruption of lower incisors in 3 pups in the high

dose group, incomplete appearance of abdominal hair in one pup each in the control and the high dose groups and incomplete descent of testes in one animal each in all three groups were observed on the last scheduled day of observation as "incomplete differentiation". However, these processes were completed by the following day. The results of the righting reflex, negative geotaxis, free-fall righting reflex, sensory function, emotionality and learning tests were similar in treated and control groups.

TABLE 3.5.6.1
PERINATAL AND LACTATIONAL PERIODS IN RATS: MEAN F_1 POSTNATAL BODY WEIGHTS

Dose, mg/kg	Sex	Body weight (in gm) on postnatal day									
		0	4	8	15	22	29	36	43	50	57
0	M	7.3[a]	10.9	21.2	40.0	66.0	115.8	180.1	244.3	301.9	354.8
	F		10.6	20.3	38.5	63.2	106.5	152.2	185.8	213.8	230.3
0.3	M	7.4[a]	10.8	20.6	39.3	65.6	115.0	180.0	243.7	301.9	353.3
	F		10.4	19.8	37.8	62.6	105.2	150.9	185.0	211.9	236.6
1.6	M	7.1[a]	10.1§	19.9*	38.2*	63.3§	111.4*	175.8*	238.4*	296.0*	348.3*
	F		9.8§	19.2§	37.0§	60.7§	101.5*	146.2*	182.1§	208.5*	232.7*

a: sexes combined
* $p < 0.01$ compared with control
§ $p < 0.05$ compared with control (calculated by the reviewer using the Student's t-test)

OM had no adverse effects on the reproductive capacity of the F_1 animals. All F_1 dams showed normal parturition and no significant difference from control in number of newborns and mean birth weight. No abnormalities in external appearances were noted in F_2 pups. Autopsy of F_1 animals used for the reproductive test showed no macroscopic abnormality of the thoracic/peritoneal viscera in any of the groups. Autopsy of F_1 animals not used for the reproductive test revealed dilatation of the renal pelvis in all groups (incidence in treated groups similar to control group incidence). No abnormalities were detected in epididymal sperm.

In summary, olmesartan medoxomil administration during late gestation and lactation had no adverse effects on maternal body weight gain, food intake or parturition. A continuous suppression in body weight gain was observed in F_1 animals in the high dose group post weaning. Timing of postnatal maturation did not differ between treated and control groups. The previous study (section 3.5.5) documented a significant increase in the incidence of dilatation of the renal pelvis in the F_1 generation at doses of 8 or more mg/kg/day. In contrast, in the present study, dilatation of the renal pelvis was observed in a few cases in all groups including the control group ($p > 0.05$) and thus, was not considered attributable to the treatment with OM. Based on these results, the no effect dose level for abnormal development of the next generation in rats is 0.3 mg/kg/day.

4. OVERALL SUMMARY AND EVALUATION

Pharmacodynamics

Olmesartan medoxomil (OM) is a non-peptidic, orally effective, potent and specific antagonist of angiotensin II, active at the AT_1 receptor. It was developed by Sankyo Pharma Inc., for the treatment of essential hypertension.

OM is a prodrug that is rapidly hydrolyzed by aryl esterase (present in most species including human) to olmesartan. One mg OM is equivalent 0.8 mg of olmesartan. The sponsor used OM in *in vivo* studies employing oral administration (as suspension). Olmesartan was used for all *in vitro* studies and *in vivo* studies employing intravenous administration,. The potencies of OM and olmesartan were compared in receptor specific binding studies. Based on the IC_{50} values, OM has about one-fourth the AT_1 receptor antagonistic activity of olmesartan. In both receptor specific binding and isolated tissue studies, olmesartan was shown to be more potent than losartan and equal in potency to candesartan cilexetil. These studies characterized OM and olmesartan as a noncompetitive, insurmountable antagonists. This could be one of the reasons why the effects of OM are long lasting in both rats and dogs (see below). Losartan, the prototype AT_1 receptor antagonist, is a competitive, surmountable antagonist but its active metabolite, EXP-3174 is a noncompetitive (competitive at low concentrations) and insurmountable antagonist.

In normotensive rats, OM significantly decreased blood pressure (and increased heart rate) in a dose-dependent manner at doses as low as 1 mg/kg. The maximal responses occurred 4 to 7 hours after administration and blood pressure returned to pretreatment levels within 20 hr after dosing. Such activity in normotensive rats is not seen with losartan, telmisartan or irbesartan but present in candesartan cilexetil. The antihypertensive effect of OM depends on the activity of the intact renin-angiotensin system, as demonstrated by its effectiveness in inhibiting the pressor responses to infused angiotensin II and in different animal models of hypertension. In angiotensin II- induced hypertension in rats, both olmesartan (0.01 and 0,.03 mg/kg) and OM (0.03 and 0.1 mg/kg), caused dose-dependent decreases in b.p. within 30 min of administration and the inhibitory effect lasted for more than 8 hr. Similar observations were made in dogs except that the doses were 10-fold higher and the inhibitory effect had not fully recovered even 24 hr after administration.

OM was about 10 times more potent as an antihypertensive agent in renal hypertensive rats (RHR) than in spontaneously hypertensive rats (SHR) and about 100 times more potent in lowering blood pressure in SHR than in normotensive rats (Table 4.1). In both RHR and SHR, the peak activity was observed about 6 hr after administration. Daily treatment with OM (0.1, 0.3 and 1 mg/kg, p.o.) for 2 weeks in SHR decreased blood pressure in a dose-dependent manner and the maximal response was unchanged throughout the treatment period. The b.p. returned to pretreatment levels 4 days after the cessation of treatment and no tolerance developed. OM at 30 mg/kg had a negligible hypotensive effect in DOCA-salt hypertensive rats, a low-renin model of hypertension. This indicates that normal levels of plasma renin activity are necessary to demonstrate the antihypertensive action of OM.

TABLE 4.1
24-HR HYPOTENSIVE AREAS FOR OLMESARTAN MEDOXOMIL IN CONSCIOUS RENOVASCULAR HYPERTENSIVE (RHR), SPONTANEOUSLY HYPERTENSIVE (SHR), NORMOTENSIVE (NR) AND DOCA-SALT (DOCA) HYPERTENSIVE RATS

	Single Oral Dose							
	0.01 mg/kg	0.03 mg/kg	0.1 mg/kg	0.3 mg/kg	1 mg/kg	3 mg/kg	10 mg/kg	30 mg/kg
RHR	71 ± 70	248 ± 94	412 ± 95	439 ± 171				
SHR			198 ± 44	286 ± 73	357 ± 45			
NR					87 ± 27	156 ± 59	185 ± 21	245 ± 29
DOCA								56 ± 91

Values are mean ± SEM

In Goldblatt hypertensive dogs, OM significantly decreased blood pressure after oral administration (3 and 10 mg/kg/day) for 14 days. OM also significantly increased plasma renin activity and angiotensin I and II levels. It is reasoned that angiotensin II receptor antagonism removes the negative feedback on renin secretion, resulting in increased renin secretion and, consequently, increased levels of angiotensin I and II.

The beneficial effects of OM on the cardiac hypertrophy and nephropathy that develop secondary to hypertension were investigated in SHR. OM (1 mg/kg, p.o.) significantly decreased the heart to body weight ratio in SHR relative to that of vehicle treated SHR and (at 3 and 10 mg/kg, p.o.) significantly decreased the elevated levels of urinary protein excretion and urinary N-acetyl-ß-D-glucosaminidase activity, an index of tubular injury.

Hemodynamic studies conducted with olmesartan in intact dogs at doses as high as 10 mg/kg, i.v. suggest that its cardiovascular effects are limited to the hypotensive effect expected from its action at the AT_1 receptor. General pharmacology studies demonstrated little effect on a variety of physiological systems, except for those effects that would be expected based on the above noted activity. Based on these studies, it is concluded that OM would produce minimal adverse effects at pharmacological doses.

Drug Disposition (ADME)

Absorption

Olmesartan medoxomil was rapidly absorbed after oral administration (t_{max} 1/2-3 hr) to rats, dogs and humans. T_{max} tended to increase at higher doses. C_{max} and AUC were generally higher in females than in males. These values were not influenced by food in the stomach. The absolute bioavailability of olmesartan following oral administration of OM to rats was 28% at 5 mg/kg and 31% at 10 mg/kg when administered in suspension and about 74% when administered to rats at 5 mg/kg in solution. Additionally, absorption of the pro-drug, OM, in rats was approximately 3-fold higher than that of the active metabolite, olmesartan. In dogs, the bioavailability of olmesartan after oral administration of 2.3 mg/kg OM was 14% when given in solution and 12% when given in capsule. Pretreatment of dogs with pentagastrin increased bioavailability to 18%

from the solution and to 23% from the capsule. In humans, the calculated mean bioavailability of olmesartan following oral administration of a 20 mg OM tablet was 26%.

^{14}C tagged OM was rapidly eliminated (as olmesartan or radioactivity) after a single dose with half-lives ranging from 0.75 to 1.5 hr in dogs and 1 to 3.5 hr in rats. In contrast, in humans the elimination half-life ranged from 14 to 18 hr. Decreased renal function as evidenced in nephrectomized rats did not adversely affect pharmacokinetic parameters except for a slight increase in mean residence time and total clearance. Plasma concentrations of olmesartan were similar after repeated dose administration of OM, suggesting no accumulation of drug-related substances with time.

Distribution

With oral administration of ^{14}C-OM to rats, olmesartan was the main component of the plasma radioactivity (96% at 15 min). It does not penetrate red blood cells. In the male rat, absorption of radiolabeled OM from the gastrointestinal tract was rapid with peak blood and tissue levels appearing between 0.25 and 2.50 hr. High concentrations of radioactivity were observed in the liver, exceeding the plasma concentration by 2- to 5-fold for up to 8 hr after administration. The concentration of radioactivity in other tissues was in the following rank order: liver >> kidney ≥ blood, lung, skin > heart > other tissues and organs. There was no uptake into the central nervous system or eyes. The plasma concentration-time curve showed a monophasic profile with a half-life of 1.68 hr. Similarly, the liver showed a monophasic profile with a half-life of 1.55 hr. The elimination half-lives in other tissues and organs ranged from 1.5 hr to 2.7 hr, showing decreases in parallel to the decrease of the plasma concentration. The radioactivity concentrations in most of the organs and tissues were 8% and 4% of their maximal levels at 24 and 72 hr, respectively, after the last dose. By 168 hr after the last dose, radioactivity was detectable only in fat (9.2% of maximum levels), lung, kidney, skin and colon (0.2% to 2.3% of maximum levels). The concentration of radioactivity in tissues was comparable after a single dose and after 21 days of dosing. The [redacted] revealed that parent compound was almost undetectable and that olmesartan accounted for 88% and 76% of the radioactivity in the liver and kidney, respectively. The olmesartan glucuronide conjugate accounted for 1.65% and 5.23% of the radioactivity in the liver and kidney, respectively.

In pregnant rats (on day 13 or 18 of gestation), levels of radioactivity in the uterus, ovary, mammary gland, placenta, amniotic fluid and fetus, at 1 hr, were lower than the level in blood. However, 24 after dosing (on GD 18), fetal levels were higher than maternal plasma levels and higher than fetal levels 1 hr after dosing. After a single oral administration of ^{14}C-OM to lactating rats, radioactivity was detected in milk at levels lower than in plasma. Peak levels in milk occurred after approximately 4 hours, whereas peak levels in plasma occurred within 30 minutes. Mean $AUC_{(0-\infty)}$ for radioactivity in milk was about one-fourth the AUC value for plasma.

Olmesartan was highly bound to serum proteins from mice, rats, dogs, and humans (>94%). It was also highly bound to human serum albumin (>99%) and human serum α_1 acid glycoprotein (96%), but not human serum globulin (13%). Olmesartan was mainly bound to the warfarin site on human albumin.

Metabolism

The effect of OM on drug-metabolizing enzymes was studied both *in vitro* (human liver microsome fraction) and after repeated-dose oral administration to rats for 7 days. At *in vitro* concentrations corresponding to the maximal plasma concentration observed in clinical trials (10 μM), olmesartan produced no inhibition of a range of drug-metabolizing enzymes, except for an inhibition (20%) of chlorzoxazone hydroxylase (CYP2E1), thus suggesting that interactions with drugs that inhibit, induce or are metabolized by P450 enzymes other than CYP2E1 are not expected. After repeated oral administration to rats, OM significantly and dose-dependently decreased cytochrome P450 content compared to control values. According to the sponsor, this could be due to a decrease in unknown isoforms and/or may be related to a decrease in body weight gain (~12%).

OM was rapidly and completely hydrolyzed to olmesartan after oral or I.V. administration. *In vitro* studies have shown that OM is hydrolyzed by human serum albumin and by plasma esterases in mouse, rat, dog, rabbit, monkey and human plasma. The highest activity was observed with rabbit plasma (41.58 nmol/min/mg) and the lowest with rat plasma (0.97 nmol/min/mg); the activity of human plasma was 10.58 nmol/min/mg. Studies to characterize the plasma esterase responsible for hydrolysis of OM indicated that it was arylesterase. OM was also hydrolyzed by S9 fractions of rat liver and small intestine. The hydrolyzing activity of the hepatic fraction was approximately 7-fold higher than that observed with the small intestine fraction. Hydrolysis of OM is likely to proceed at the cyclic diester bond of carbonic acid in RNH-8097 and not at the ester bond between the carboxyl group of olmesartan and the hydroxyl group of RNH-8097, as no RNH-8097 was detected in any species (see Fig. 2.7.1). The formation of olmesartan glucuronide was observed only in S9 fractions of rat small intestine. Fifteen min after oral administration of [^{14}C]OM to rats, olmesartan and olmesartan glucuronide accounted for 89 and 9%, respectively, of the total radioactivity observed in plasma. OM accounted for only ~1% of the total radioactivity in the plasma. The proportion of radioactivity accounted for by olmesartan decreased as the proportion of olmesartan glucuronide increased. In contrast, in the small intestine, OM accounted for 36.4% of the total radioactivity, olmesartan accounted for 56.2% of the radioactivity, and the glucuronide accounted for 5.1% 1 hr after oral administration of [^{14}C]OM to rats. No other metabolites were observed. Bile duct occlusion and renal artery ligation had no effect on the metabolism of OM. Olmesartan was also the main metabolite observed in urine, feces and bile, again, with no OM observed. After oral, but not I.V. administration, bile contained substantial amounts of olmesartan glucuronide. This suggests the conjugation reaction occurs in the intestinal tract. No other metabolites were observed in bile. In dogs, unchanged OM was not observed in urine or feces and olmesartan accounted for all of the radioactivity. It is concluded that following the rapid and complete conversion of OM to olmesartan during absorption, there is virtually no further metabolism.

Excretion

After oral or intravenous administration of [^{14}C]OM to rats, less than 2% of the radioactivity was recovered in the urine (remainder recovered from feces). After oral administration to dogs, urinary excretion accounted for about 9% of the dose (fecal excretion about 83%) whereas following intravenous administration, about 20% was recovered in the urine (about 78% recovered from feces). Similar results were observed in humans after a single oral administration of 20 mg of [^{14}C]OM, with urinary excretion of radioactivity accounting for about 13% of the

dose and fecal excretion accounting for about 77% of the dose. In all species examined (including human) more than 98% of the radioactivity in fecal samples was accounted for by olmesartan. The high level of fecal recovery following oral or intravenous administration to rats was attributed to biliary excretion. A biliary excretion study in rats showed that 24 hr following oral or intravenous administration of [^{14}C]OM, most of the radioactivity had been excreted in bile (as olmesartan and olmesartan glucuronide after oral administration; as olmesartan after intravenous administration).

Toxicology

Acute Toxicity

Single dose oral toxicity studies were performed with olmesartan medoxomil in mice, rats and dogs. No clinical signs were observed at the highest dose tested (2000 mg/kg in the rodents and 1500 mg/kg in dogs).

Chronic Toxicity

Rats:

The chronic toxicity of olmesartan medoxomil (OM) was evaluated in rats in gavaging studies at doses of up to 1000 mg/kg/day for 6 months and in a dietary administration study at doses up to 2000 mg/kg/day for 24 months (carcinogenicity study). In these studies, there were no remarkable treatment-related clinical findings and olmesartan medoxomil had no effect on survival. Significant and dose-dependent decreases in body weight gain and food consumption were observed for males at doses of 100 or more mg/kg/day for the first 3 weeks of dosing in the 6 month study. Red blood cell indices (RBC, hemoglobin, hematocrit) decreased (3 to 14%) significantly and dose-dependently after 3 or 6 months at 30 or more mg/kg/day in both sexes. However, mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration increased significantly at doses as low as 30 mg/kg/day in females and 300 or more mg/kg/day in males at these same intervals. Significant and dose-dependent increases in platelet counts (9-11%) were noted in females after 6 months at 300 or more mg/kg/day. Males given 300 or more mg/kg/day for 6 months showed significant increases in percentage of neutrophils (66-77%) and significant decreases in percentage of lymphocytes (12%). Blood chemistry examinations showed a significant increase in BUN in males (30-60%) after 3 months and 6 months and females (18-24%) after 6 months at 100 or more mg/kg/day. Creatinine levels were increased in males (10%) after 6 months at 1000 mg/kg/day. Increases in potassium and decreases in sodium were observed in males after 6 months at 100 or more mg/kg/day. Significant decreases in AST, ALT and total protein levels were observed in all treated male groups in the 6 month study.

Organ weight analysis showed significantly reduced absolute (11 to 17%) and relative (10 to 15%) heart weights in both sexes at doses as low as 10 mg/kg/day administered for as little as 3 months. This is an effect that has been observed with other AT-1 receptor antagonists as well as ACE inhibitors. Absolute and relative kidney weights were significantly increased for males after 6 months at 100 or more mg/kg/day (13 to 16% and 9 to 13%, respectively). For females, high absolute and relative weights of kidneys (6 to 8% above control) were observed at doses as low as 30 mg/kg/day in the 6 month study. Furthermore, absolute and relative adrenal weights were significantly increased (14 and 17%, respectively) in males receiving 1000 mg/kg/day. The

kidney was the target organ for toxicity. Progressive nephropathy was observed in approximately 30% of the males treated with 300 or more mg olmesartan/kg/day in the 6 month study (seen as early as 3 months in a study in which 300 mg/kg/day was the highest dose). No degeneration or regeneration of tubular epithelium and no dilatation of the tubules was observed in the treated female groups. Thickening of arterial wall from interlobular artery to afferent arteriole was observed with greater than control frequency in all groups of OM-treated females and in males given 100 or more mg OM/kg/day. The juxtaglomerular index (JGI) was statistically significantly increased in both males and females in the 30, 100 and 300 mg/kg/day groups; values were increased in both sexes of the 1000 mg/kg/day group but not significantly ($p > 0.05$). This finding is consistent with the pharmacological site of action of the test substance as an angiotensin II receptor blocker. In the spleen, increased hemosiderin deposition was observed in males receiving 1000 mg/kg/day for 6 months. No histomorphological correlate of the decreased heart weight was noted. Toxicokinetic evaluations showed no drug accumulation after 3 or 6 months in rats given 300 or 1000 mg/kg/day, respectively.

In the 24-month carcinogenicity study, dietary administration of OM at dose levels of up to 2000 mg/kg/day elicited no clinical signs of toxicity and did not adversely affect survival. The average achieved doses of test substance were between 98.8 and 101.6% of the targeted doses. Body weights were statistically significantly lower than concurrent control for all OM treated groups, beginning on day 8 of drug administration and continuing to termination of the study (3 to 17% lower for males, 2 to 12% lower for females). Changes in hematological parameters noticed previously in 3 and 6 month studies were not evident in this study though plasma concentrations were comparable. Statistically significant and dose-related increases in absolute and relative thyroid and adrenal weights and decreases in absolute liver weights relative to the control group were observed for male rats. However, decreases in heart weight observed in the 6 month study were not noted in the 24 month study. At necropsy, no increase in incidence of masses or nodules was noted for rats treated with OM relative to control. Histopathology considered to be related to treatment was seen in the kidneys and was similar to that observed in the 3 and 6 month studies. Additionally, increased incidences of esophagectasis (females), foreign body pneumonia (males and females), exudative inflammation of the eye (male), increased foci of cytoplasmic vacuolation in the adrenal glands (females) and periarteritis in the testis (males) were noted at the highest dose of the test substance. Both the sponsor's and the agency's analyses revealed no statistically significant increased trend in the incidence of any neoplasm that could be attributed to treatment with OM for rats of either sex that survived the treatment period or that were killed or died during the treatment period. A nominal increase in the incidence of renal tubular neoplasia in treated males (adenomas and carcinomas which were not seen in the concurrent control rats, and are relatively rare on the basis of historical control data for the strain (0.8%)) was not statistically significant or dose-dependent. Mean plasma concentrations of olmesartan increased with increasing dose in both sexes. The 2000 mg/kg/day treatment in male rats was associated with a mean plasma concentration (5.8 µg/ml) 8-fold higher than the mean C_{max} in male humans after 10 daily oral doses of 40 mg (0.73 µg/ml).

The Executive Carcinogenicity Assessment Committee (CAC) at its meeting on March 20, 2001 expressed concerns about the occurrence of renal tubular cell tumors and renal tubular hyperplasia in only OM-treated animals. The full CAC was asked to assess the evidence of carcinogenic potential for OM in the rat (as well as the adequacy of both the p53 (+/-) and Hras2

assays in the mouse). A full CAC meeting was held on May 4, 2001 and the participants included representatives and consultants for Sankyo Pharma. Twelve of the 20 CAC members felt that the drug had tested positive in the rat carcinogenicity assay, with 8 of these 12 saying that they could not conclude that the finding would have little or no relevance to human cancer risk. Seven of the 20 asked that a third party chosen by the agency redo the histopathology. An additional meeting was held with the sponsor on May 30th. At that meeting, the agency asked the sponsor to do additional evaluations to demonstrate that OM is not a carcinogen. In this context, the agency suggested that a kidney step-sectioning protocol be carried out and that the original study kidney slides be sent to NIEHS for peer review.

The peer review by NIEH pathologists and an independent expert review by [redacted] did not significantly alter the tumor findings of the original study pathologist. The original diagnosis of tubule hyperplasia was not confirmed by [redacted], but rather, was found by him "to represent an adaptive change of renal tubule hypertrophy". The NIEH pathologists concluded that the tubular hyperplasia observed in the original study reflects tubular regeneration secondary to nephropathy and that it is unlikely that these lesions are part of the neoplastic process. PCNA (proliferating cell nuclear antigen) staining further corroborated the lack of a drug-induced proliferative lesion. The step-section analysis of the kidneys resulted in four more tumor-bearing animals in comparison with the original (single section) evaluation. After step-sectioning, the number of tumor bearing animals was 0, 3, 5 and 4 in the control, low, mid and high dose groups, respectively. Though the number of tumor bearing animals in the step-section analysis increased from 8 to 12, the sponsor still contends that the incidence is not significantly different from the concurrent control group incidence (0%) and that the incidence for each treated group is within the range of historical control group values for the step-section protocol (mean incidence, 4.62%; range, [redacted] %). The FDA analysis of the step-section data revealed no statistically significant increased trend in the incidence of adenomas and carcinomas that could be attributed to treatment with OM ($p=0.12$). But, pair-wise comparisons (mortality adjusted or unadjusted) resulted in a statistically significant differences between control and the mid dose group ($p=0.02$). Furthermore, control versus all treated males (combined) resulted in a p-value of ≤ 0.02.

Mice:
The carcinogenic potential of olmesartan medoxomil was evaluated in 26 week studies in two strains of mice (alternatives to the standard 2 year carcinogenicity bioassay). The first study was conducted in the strain C57BL/6TacfBR-[KO]N5 p53(+/-) with oral (gavage) doses of up to 1000 mg/kg/day. There were no drug-related clinical signs, unscheduled deaths or effects on body weight. There were no apparent effects on hematology parameters or organ weights. Drug-related histopathology, consistent with that seen in other repeat dose toxicity studies, was observed in the kidneys. Malignant lymphoma (lymphocytic) occurred in one female in each of the treated groups. Granulocytic leukemia was noted in a low dose female and a mid dose male. Thus, hematological neoplasia was present in all treated groups (2, 2 and 1 in animals receiving 100, 300 and 1000 mg/kg/day, respectively). There were no similar findings in the concurrent vehicle control group. However, several published studies report incidences of granulocytic leukemia and malignant lymphoma in untreated p53(+/-) and wild type mice (+/+). Neither the sponsor's nor the Center's statistician found a significant increasing trend for hemato neoplasia ($p>0.05$). Thus, it may be concluded that OM exhibited no carcinogenic potential in this alternative mouse model at a dose of 1000 mg/kg/day. In the positive control group, treated with

p-cresidine, there was a single transitional cell carcinoma of the urinary bladder in one male and hyperplasia of the transitional epithelium of the urinary bladder in all other animals, the latter finding said to be predictive of ultimate bladder tumorigenicity in these animals. The pathology was more severe in males than in females. However, the study failed to demonstrate a statistically significant positive tumor response in the *p*-cresidine exposed mice. The reason for this failure could be differences in the method of administration of *p*-cresidine. After reviewing the data from several studies, Storer observes that *p*-cresidine appears to be working reliably as a positive control compound for induction of bladder tumors with dietary administration at dose levels from 2500 to 5000 ppm producing higher incidences of lesions than produced by daily gavage dosing in corn oil at 400 mg/kg (Storer, R: *The p53 workgroup newsletter*, Vol. 1 (1), August 1998, Merck & Co., Inc. West Point, PA). Plasma concentrations of olmesartan showed a non-proportional increase over the dose range studied. The 1000 mg/kg/day treatment in male p53(+/-) mice was associated with systemic exposure (mean concentration determined 2 hr post dose at 26 weeks, 3.23 µg/ml) 4.5-fold higher than the C_{max} in male humans after 10 oral daily doses of 40 mg (0.73 µg/ml).

The supplemental 26-week **non-GLP** carcinogenicity study used the transgenic mouse strain, CB6F1-TgHras2. Carcinogenic potential of olmesartan medoxomil (OM) was evaluated with dietary administration at a single dose level of 1000 mg/kg/day. Vehicle (corn oil by gavage) and positive control (i.p., N-methyl-N-nitrosourea) groups received their diet in pellet form whereas the test group animals were fed a powdered diet. There were no drug-related clinical signs or unscheduled deaths. Unlike the p53(+/-) mouse study, mean body weights of males treated with OM were significantly lower (4 to 9%) than control. Red blood cell indices were also significantly lower than control (8 to 14%) whereas relative increases were seen in BUN (45 to 86%), creatinine (13 to 20%), alkaline phosphatase (15-20%) and transaminases (14-27%, only in males). Significantly lower than control absolute and relative heart and thymus weights and higher than control absolute and relative liver weights were observed in OM-treated animals of both sexes. Furthermore, absolute and relative adrenal weights for OM-treated males were significantly lower than control whereas absolute and relative kidney weights for OM-treated females were significantly higher than control. The OM-treated animals exhibited JG cell hyperplasia in the kidneys, which had been observed previously in rats and dogs. There were no significant differences in the incidence of neoplastic lesions between the drug treated group and the vehicle control group. Positive control mice were diagnosed with malignant lymphoma, squamous cell papilloma and carcinoma in the forestomach, and hemangio-tumors. Based on mean AUC values, the systemic exposure at the dose of 1000 mg/kg/day in the Hras2 transgenic male mouse (determined at 26 week, 47.4 µg.h/ml) exceeds the human AUC (10 daily oral doses of 40 mg olmesartan in male volunteers, 4.4 µg.h/ml) by a factor of 10.9.

Majority of the CAC members at its meeting (as referenced above) felt that the p53 assay was probably an adequate test of carcinogenicity. Many had concerns about the lack of tumors in the positive control, suggesting that this reflected either a failure or a low sensitivity of the assay and perhaps a capability of detecting only potent carcinogens. Regarding Hras2 assay, 19 of 20 members said that the test dose administered was adequate as it reached the maximum tolerated dose.

Dogs:
The chronic toxicity of olmesartan medoxomil was evaluated at oral doses of up to 500 mg/kg/day for 3 months and 160 mg/kg/day for 1 year. The following findings were associated with chronic exposure to the drug: excretion of white material in the feces (indicating excretion of unchanged drug substance) at 500 mg/kg/day and slight anemia at 125 or more mg/kg/day (as early as 3 months). Blood urea nitrogen and creatinine were elevated moderately (less than 2-fold) and mild dilatation of renal tubules with mild to moderate regeneration of tubular epithelium (suggesting occurrence of tubular damage) was observed at doses as low as 125 mg/kg/day (only in 1 of 6 animals at this dose) in the 3 month study; at 500 mg/kg/day one of six animals had to be sacrificed in moribund condition (in renal failure) after about 1 month of treatment. Similar associations with treatment were not observed in the 1 year study at 160 mg/kg/day. The reason for this apparent discrepancy is not clear. Both Cmax and AUC values at 125 mg/kg/day given for 3 months (1.18 to 3.21 µg/ml and 7.13 to 24.66 µg.hr/ml, respectively) are comparable to values at 160 mg/kg/day given for 1 year (1.20 to 1.82 µg/ml and 7.26 to 15.41 µg.hr/ml, respectively). The male dog receiving 125 mg/kg/day that exhibited regeneration of tubular epithelium had AUC values of 15.35 and 16.99 µg.hr/ml, on days 28 and 91, respectively. The high dose dog sacrificed in moribund condition after about 1 month into treatment had an AUC value of 21.2 µg.hr/ml on day 28. The exaggerated pharmacological effect, manifested as JGA hyperplasia and a mild increase in granules in juxtaglomerular cells, was observed at doses of 125 or more mg/kg/day in both the 3 and 12 month studies. Both AUC and C_{max} values increased, in a less than proportional manner, as the dose increased to 125 or 160 mg/kg/day in the 3 month or 12 month study, respectively. Higher doses resulted in either a small increase, a small decline or a plateau. In both studies, AUC and C_{max} values were not measurably changed after repeated administration, suggesting that there is no olmesartan (active metabolite) accumulation at these dose levels. The 160 mg/kg/day treatment was associated with systemic exposures that were, on average, only twice as high as those observed in healthy male volunteers at the maximum recommended daily dose of 40 mg. However, it should be noted that oral administration of olmesartan medoxomil to dogs at a dose of only 1 mg/kg produced an 87% inhibition of the angiotensin-II (0.01 mg/kg, i.v.)-induced pressor response.

Genotoxicity

An extensive investigation for evidence of genetic toxicity was conducted with olmesartan medoxomil and its metabolites. A battery of *in vitro* studies was conducted –including the Ames reverse mutation assay, the Chinese hamster lung cell chromosomal aberration assay, the mouse lymphoma cell assay and the Syrian hamster embryo cell transformation assay. In vivo studies included the mouse micronucleus test, an unscheduled DNA repair test in rat hepatocytes, gene mutation assays in transgenic mice (Muta™ mouse intestine and kidney), and comet assay to detect DNA damage in the kidney.

Olmesartan medoxomil (OM), olmesartan, HMPIC, MBT and acetoin were negative in all tester strains in the Ames reverse mutation assays both in the presence and absence of S-9 mix. In contrast, RNH-8097 produced a dose-dependent and marked (4.9-fold higher than vehicle control) response in TA100 and was weakly positive in TA1537 and WP2uvrA, all in the absence (but not in the presence) of S-9, and diacetyl was positive in strains TA100 and

WP2uvrA (more than 2-fold higher relative to vehicle control) in the presence (but not in the absence) of S-9 mix (see summary Table 4.2).

TABLE 4.2
SUMMARY OF GENETIC TOXICOLOGY DATA FOR OLMESARTAN MEDOXOMIL AND ITS METABOLITES

Compounds tested	Ames assay[1]		CA[2]		TK[3]		SHE[4]	MN[5]	DNA[6]	TG Mouse[7]		Comet assay[8]	
	-S9	+S9	-S9	+S9	-S9	+S9				int	kid	Y	O
OM	-	-	+	-	+	+	-	-	-	±[9]	-	?[10]	-
Olmesartan	-	-	-	+	NT	NT	-	-	NT	NT	NT	NT	NT
RNH 8097	+	-	-	-	NT	NT	NT	NT	NT	NT	NT	NT	NT
Diacetyl	-	+	+	+	NT	NT	NT	NT	NT	NT	NT	NT	NT
HMPIC, MBT, Acetoin	-	-	-	-	NT	NT	NT	NT	NT	NT	NT	NT	NT

Responses are shown as: - negative, + positive
NT: not tested, int: intestine, kid: kidney, Y: young rats, 9 week old, O: 9 month old

1: Reverse mutation in bacteria
2: *In vitro* chromosome damage
3: *In vitro* gene mutation, mouse lymphoma assay (TK locus)
4: Morphological cell transformation
5: *In vivo* micronucleus test
6: *In vivo* Rat hepatocyte UDS
7: *In vivo* gene mutation assay in transgenic mice (Muta™ mouse assay), evaluation in intestine and kidney
8: *In vivo* study of DNA migration in kidneys of young and old rats
9: Of the three studies, first two were positive and the third negative for effects on the *LacZ* gene. The third study was positive for effects on the *CII* gene.
10: Differences in the analysis of data between the agency (positive finding) and the sponsor (negative finding). See appendix 1 for details.

Two *in vitro* chromosomal aberration assays were conducted with OM in Chinese hamster lung cells. In both studies, OM tested positive in the absence (but not in the presence) of S-9 mix, inducing dose-dependent chromatid gaps, breaks and exchanges. However, there was no evidence of significant induction of chromosome breaks or exchanges. With DMSO used as vehicle, the active metabolite, olmesartan, tested negative. With CMC as the vehicle, olmesartan at 0.0781 mM but not 0.156 mM was associated with numerical aberrations (6-11% of cells) in the absence of S-9. Absence of a dose-response relationship and strong cytotoxicity suggests a non-specific response. On the other hand, structural aberrations (22%; chromatid breaks and exchanges and chromosome exchanges) were found at a high concentration (10 mM), with no cytotoxicity, in the presence of S-9 mix. Diacetyl (in CMC vehicle) but not RNH-8097, tested positive in both the presence and absence of S-9 mix. The effects of diacetyl were limited to chromatid breaks and exchanges. HMPIC, MBT and acetoin (all in CMC vehicle) did not produce structural or numerical aberrations. These results suggest that olmesartan and diacetyl are responsible for the chromosomal aberrations induced by OM.

The differing results with OM in the presence *versus* the absence of S-9 in these studies could be explained on the basis of the *in vitro* conversion of OM to olmesartan and diacetyl. In the absence of S-9, OM should pass the cell membrane and be almost completely hydrolyzed to olmesartan and diacetyl in the cytoplasm (Fig. 4.1, top). Thus, the nucleus is exposed to a high concentration of diacetyl, which results in chromosome aberration. In the presence of S-9, OM is mostly hydrolyzed to olmesartan and diacetyl in the culture medium. Then S-9 further metabolizes diacetyl to acetoin or 2,3-butanediol, neither of which is clastogenic (Fig. 4.1, bottom). Both olmesartan and diacetyl produce a positive response in the presence of S-9, suggesting that large amounts of non-hydrolyzed olmesartan and diacetyl need to pass the cell membrane for those compounds to be effective clastogens. This is supported by the experimental observation that diacetyl tested positive at 1 and 10 mM in the absence and presence of S-9, respectively.

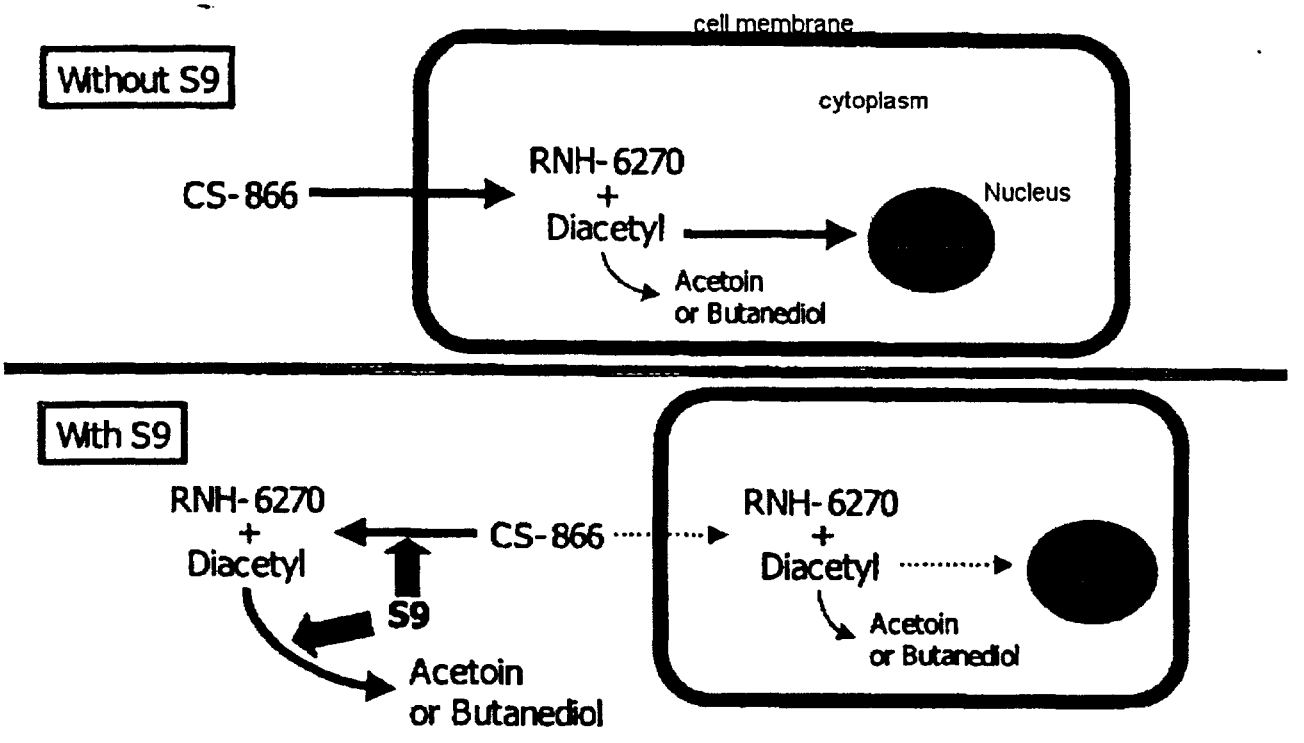

Fig. 4.1. *In vitro* kinetics of olmesartan medoxomil

In the mouse lymphoma cell assay, OM induced dose-related increases in TK mutant frequency in both the absence and presence of S-9 mix. The responses were statistically significant at moderate levels of toxicity (20-45% relative survival). OM induced both large and small colonies. According to Applegate *et al.* (1990)[1], induction of small colony mutation is frequently indicative of large (intergenic) lesions in the DNA, characteristic of chromosome-type damage. The large colonies most likely represent intragenic mutations caused by missense events and microdeletions or additions of a few DNA base pairs. Olmesartan and the other metabolites of OM were not directly tested in the mouse lymphoma cell assay.

[1] Applegate, M.L. *et al.* (1990): Molecular dissection of mutations at the heterozygous thymidine kinase locus in mouse lymphoma cells. *Proc Natl Acad Sci.*, 87: 51-55.

OM and olmesartan tested negative in the *in vitro* Syrian hamster embryo cell transformation assay. Diacetyl and RNH 8097 were not tested in this assay system.

The mouse micronucleus and rat unscheduled DNA repair tests failed to demonstrate a potential for mutagenicity of OM *in vivo*. Olmesartan and the other metabolites of OM were not directly tested.

Since OM is intended for oral dosing and is metabolized in the intestine, the intestine was examined as a potential target organ for genetic toxicity in transgenic mice (Muta™ mouse assay). In two studies, small (less than 2-fold relative to vehicle control) but statistically significant increases in *lacZ* mutation frequency were observed in the intestinal epithelium from all OM-treated groups (100, 1000 or 2000 mg/kg/day p.o. for 5 days). The increase in MF for OM-treated groups is questionable since the response was not dose-dependent and less than 2-fold relative to vehicle control. Additionally, the positive control, dimethyl hydrazine, showed a minimally sufficient response (1.9 to 2.4 times vehicle control). Thus, the sponsor repeated the Muta™ mouse assay with the intestine (evaluated both *lacZ* or *cII* mutation frequencies) and also conducted a Muta™ mouse assay with the kidney (only *lacZ* mutation frequency). In both studies (reports submitted as amendments to the NDA), no statistically significant increases in *lacZ* mutations in the intestine and kidney relative to vehicle control were observed 1 or 14 days after 5 days of OM administration. In contrast, OM tested positive for *cII* gene mutations in the intestine 14 days after 5 days of OM administration The response to the positive control was remarkably high with the intestine (25 to 35 times vehicle control with MNU) but not with the kidney (2.4 times vehicle control with STZ). If the criterion for a positive study is a mutation frequency at least 2 times vehicle control, then OM is not mutagenic either to the intestine or to the kidney.

A comet assay, which detects DNA damage, was carried out in (9 weeks old) rats of the strain employed in the 2 year carcinogenicity study, and the effects on kidney DNA evaluated. At the present time, because of variations in analyses of data between the sponsor and the Center's genetic toxicology committee (see appendix 1 for details), the results of the study are considered equivocal. The sponsor calculated the DNA migration for each animal by averaging 50 migrations. They then compared the mean group values of the OM-treatment and vehicle control groups. The sponsor concludes that the DNA migration (indicative of DNA damage) in animals treated with OM (600 or 2000 mg/kg) did not increase in comparison with the corresponding vehicle control group. But the genetic toxicology committee, which reviewed the results of the assay, argues that "because most of the cells in the mixed population are undamaged, and a very large effect in the majority of cells would be required to shift the mean, the proportion of damaged cells should also be analyzed." According to the Committee's analysis, OM induced damage in rat kidney at both dose levels ($p < 0.05$). In contrast, a second comet assay using 9 month old rats showed no increase ($P > 0.05$) in DNA damage with OM in comparison with the corresponding vehicle control. The results of that second study suggest that spontaneous DNA damage increases with age. The Committee thought that a high background in that second study might mask the effect that had been noted in younger animals. The genetic toxicology committee concurs with the sponsor's interpretation of the data in aged rats but does not feel that the results of that study negate their interpretation of the results of the earlier study (see appendix 2 for details).

In conclusion, OM was shown to induce chromosomal aberrations and TK mutations in cultured cells *in vitro*. Equivocal and negative results, respectively, were obtained for the active metabolite, olmesartan, in an *in vitro* chromosome aberration assay and an Ames test (not tested in mouse lymphoma assay). Diacetyl, a side-chain cleaved by ester hydrolysis during the process of absorption from the gut, produced positive responses in both Ames and *in vitro* chromosomal aberration assays. The latter result is consistent with findings in the open literature. It should be noted that diacetyl occurs naturally in foods and drinks and is categorized as "Generally Regarded As Safe (GRAS)" by the FDA[2] and as "Category A: flavoring substances which may be used in food stuffs" by Council of Europe's Committee of Experts on Flavoring Substances[3] and is not considered to pose a carcinogenic risk to humans. Based on the diacetyl content in various foods and drinks as reported in the literature, the sponsor estimates the daily intake of diacetyl in humans from food and drink to be more than 10 mg. This amount is larger than the amount of diacetyl (1.5 mg) produced from a maximum recommended daily dose of 40 mg OM (assuming 25% bioavailability).

Additionally, OM tested positive for *CII* gene mutations in the intestine and negative for *LacZ* mutations in the kidney of the MutaMouse. Equivocal results were obtained for effects on the *LacZ* gene in the intestine of the MutaMouse and in a test for DNA damage in the rat kidney (Comet assay). Olmesartan was not evaluated in the MutaMouse, micronucleus or Comet assays.

Clastogenicity and mutagenicity with olmesartan medoxomil were dose-related and the lowest concentration at which these responses were seen (124 µg olmesartan/ml, 10 µM) is much higher than encountered in human plasma (0.71 µg olmesartan/ml), suggesting that olmesartan medoxomil may pose little risk as a mutagen.

An Expert Panel constituted by the sponsor compared the genetic toxicology profile of olmesartan to that of two other angiotensin II receptor antagonists (losartan and candesartan). Based on the composite responses for these products, the panel came to the conclusion that the profile for olmesartan does not differ from that of the therapeutic class. In this reviewer's opinion, the comparison is ill founded as the results are from the limited studies done in the sponsor's laboratory. FDA reviews of reports on all of the approved angiotensin II receptor antagonists clearly show that this is not a class response but an isolated case (Table 4.3). The panel argues that the two assays that are of greatest value in defining hazard are the reverse mutation assay and the rodent micronucleus assay, noting that the "cytogenetic and mouse lymphoma tests are probably more susceptible to secondary toxicity and give a greater incidence of false positive responses."

[2] CFR report, Title 21, Vol 3, section 184, p 477-478, 1983.
[3] Flavoring substances and natural sources of flavoring. Vol 1. Chemically defined flavoring substances, Council of Europe, Maisonneuve, Strasbourg, 1992.

TABLE 4.3
COMPARISON OF THE GENETIC TOXICOLOGY PROFILE OF OLMESARTAN MEDOXOMIL WITH THE PROFILES OF DRUGS FROM THE SAME THERAPEUTIC CLASS

Assay	FDA reviews of reports on						Sponsor studies on					
	LO	VA	EP	IB	CA	TL	LO	CA	OM	OLM	RNH 8097	Di-acetyl
Reverse mutation in bacteria	-	-	-	-	-	-	NC	NC	-	-	-	+
In vitro chromosome damage	-[1]	-[1]	±[2]	-[2]	+[3]	-[2]	NC	+[3]	+[3]	+[3]	+[3]	+[3]
In vitro mammalian cell gene mutation	-[4]	-[4]	-[5]	-[4]	-[5,6]	-[4]	+[5]	-[5]	+[5]	NC	NC	NC
Morphological cell transformation (SHE)	NC	NC	NC	NC	NC	NC	NC	+	-	-	NC	NC
In vivo Rat hepatocyte UDS	NC	NC	NC	-	-	NC	NC	NC	-	NC	NC	NC
In vivo micronucleus test	-	-	-	-	-	-	NC	NC	-	NC	NC	NC
In vivo gene mutation, transgenic mice (Muta™ mouse): intestine	NC	NC	NC	NC	NC	NC	NC	-	±[7]	NC	NC	NC
In vivo gene mutation, transgenic mice (Muta™ mouse): kidney	NC	NC	NC	NC	NC	NC	NC	NC	-	NC	NC	NC
In vivo DNA damage in the kidney of 9 wk old rats (comet assay)	NC	NC	NC	NC	NC	NC	NC	NC	?[8]	NC	NC	NC
In vivo DNA damage in the kidney of 9 month old rats (comet assay)	NC	NC	NC	NC	NC	NC	NC	NC	-	NC	NC	NC

LO: losartan, VA: valsartan, EP: eprosartan, IB: irbesartan, CA: candesartan cilexetil, TL: telmisartan, OM: olmesartan medoxomil, Olm: olmesartan
NC: not conducted
Responses are shown as: - negative, + positive, ± equivocal
1: Chinese hamster ovary
2: Human lymphocytes
3: Chinese hamster lung
4: Chinese hamster V79 (HGPRT locus)
5: Mouse lymphoma assay (TK locus)
6: Chinese hamster ovary (HGPRT locus)
7: Of the three studies, the first two were positive and the third negative for effects on the *LacZ* gene. The third study was positive for effects on the *CII* gene (only one of these studies in which effect on the *CII* gene was evaluated).
8: Differences in the analysis of data between the agency (positive finding) and the sponsor (negative finding). See appendix 1 for details.

Reprotoxicity

Administration of olmesartan medoxomil (up to 1000 mg/kg/day) to male and female rats (beginning 9 weeks and 2 weeks prior to mating, respectively), had no effect on mating or fertility indices or on reproductive performance of pregnant F_0 females or on F_1 fetuses. However, mean body weight gain was statistically ($p < 0.01$) if not dose-dependently decreased (3-11%) in all male groups receiving 40 or more mg/kg/day. These groups consumed significantly less food than the control males (8-16%, $p < 0.01$) on several occasions during the treatment period.

Table 4.4 summarizes olmesartan medoxomil dosage thresholds for adverse effects observed in all rat and rabbit reprotoxicity studies other than effects in F_0 males in the fertility study.

Administration of OM to female rats (up to 1000 mg/kg/day) from 2 weeks prior to mating until day 20 of gestation did not produce any deaths or clinical signs. A statistically significant reduction in body weight gain was observed for females receiving 1000 mg/kg/day 7 days before mating (5%) and nondose-dependently at doses of 40 or more mg/kg/day between days 0 and 20 of pregnancy (6-12%). Sporadic but significant ($p < 0.01$) decreases (9-21%) in food consumption were observed in females receiving 40 or more mg/kg/day between day 14 of mating and day 7 of gestation. Treatment did not produce adverse effects on fertility or reproductive performance. Though statistically significant and dose-dependent decreases in the number of corpora lutea were observed in all treated groups (40 or more mg/kg/day) relative to the control group, number of implantations, post-implantation losses or number of surviving fetuses were not affected. It is noted that the mean corpora lutea count in the concurrent control group was outside (higher than) the laboratory historical control range. There was no effect on F_1 fetuses as assessed by body weight, external, skeletal and visceral examinations. Thus, the study suggests that OM does not have adverse effects on the F_1 generation when given to the F_0 generation before conception and during the early stages of pregnancy.

OM, at doses up to 1000 mg/kg/day, did not affect fetal survival or development when administered to rats during organogenesis (days 7 to 17 of gestation) in one study but in another (same dosage levels and days of treatment) there was a dose-dependent decrease in mean fetal weight and a dose-dependent increase in incidence of dilatation of the renal pelvis of surviving fetuses at 200 or more mg/kg/day and a decrease in the number of ossified caudal vertebrae in surviving fetuses at 1000 mg/kg/day. In both of these studies, there was decreased food consumption by dams at doses of 200 or more mg/kg/day but only in the study in which there was no evidence of adverse fetal effect was the decrease accompanied by a decrease in body weight gain. As previously noted, when female rats were treated with OM from 2 weeks prior to mating to day 20 of gestation at doses up to 1000 mg/kg/day, all doses were associated with body weight and food intake below concurrent control values but none were associated with evidence of developmental toxicity.

When pregnant rats exposed to OM (40, 200 or 1000 mg/kg/day) during organogenesis (days 7 to 17 of gestation) were allowed to deliver naturally and to rear their offspring, 3 high dose dams died several days after cessation of treatment (all during late gestation, one during labor) and 1 high dose and 1 mid dose dam died 11 days after giving birth. The high dose group had a lower

than control mean body weight (but not lower than control food consumption) during the lactation period. OM did not affect the number of newborn but a dose-dependent decrease in mean birth weight was observed in pups that had been exposed *in utero* to doses of 200 or more mg/kg/day. Exposure to OM was also associated with lower than control neonatal survival (by lactation day 4) and weight gain (body weight lower on pp day 57 than on pp day 4) at all doses.

A perinatal/postnatal toxicity study in which OM, at doses up 200 mg/kg/day, was administered from gestation day 17 through lactation day 21 demonstrated an inhibitory effect of OM on postnatal development. Statistically significant treatment-related reductions in birth weight were noted, as were significant reductions in weight gain throughout lactation and continuing post weaning for pups born to rats receiving doses as low as 8 mg OM/kg/day. There was a delay in the mean day of occurrence of most developmental landmarks (separation of ear auricula, eruption of lower incisors, appearance of abdominal hair, separation of eyelids, descent of testes and opening of vagina) in the 8 or more mg/kg/day treatment groups. Additionally, pups in the 40 or more mg/kg/day groups were slow (compared to control group pups) to respond to various exercise tests. This is probably attributable to the effect on pup body weight and relative immaturity of affected animals. Late intrauterine exposure to OM at parental doses as low as 8 mg/kg/day, with exposure continued during the suckling period, resulted in dilatation of the renal pelvis (a variation rather than a malformation) in F_1 animals. Additionally, cystic kidney was observed in a few F_1 animals in the 40 and 200 mg/kg/day groups. None of the above effects were observed in F_1 pups exposed to OM (up to 1000 mg/kg/day) only during organogenesis (days 7 to 17 of gestation). The association between the critical period of exposure beyond the second trimester, the onset of development of the RAS in the rat fetus on approximately gestation day 17, the increase in maternal and fetal exposure to OM in late gestation, and the demonstrable exposure to olmesartan during lactation, would suggest that the observed adverse fetal and neonatal effects are pharmacologically mediated. Similar observations have been made with other AT_1 receptor antagonists as well as with angiotensin converting enzyme inhibitors.

OM, like other drugs acting on the RAS, produced marked toxicity in the pregnant rabbit. Maternal toxicity resulting in death (7 of 25 does) occurred at 1 mg/kg/day. This dose also resulted in decreased weights of surviving fetuses but was not associated with adverse effects on the development of the fetal skeleton or viscera.

As with other drugs acting on the RAS, the critical periods for adverse developmental effects in the rat appear to be late gestation and lactation. Neonatal weight gain was adversely affected at doses as low as 1.6 mg/kg/day (NOEL 0.3 mg/kg/day, only about 0.07 times the maximum recommended human dose (MRHD) of OM (40 mg/day) on a mg/m^2 basis. Delays in developmental landmarks and increased incidence of renal pelvic dilatation were seen at doses as low as 8 mg/kg/day (NOEL 1.6 mg/kg/day, only about 0.4 times the MRHD of OM on a mg/m^2 basis. No teratogenic potential was observed with OM either in rats or in rabbits. However, whereas the maximum dose evaluated for teratogenic potential in rats (1000 mg/kg/day) is, on a mg/m^2 basis, about 240 times the MRHD of OM (40 mg/day), the maximum dose that could be evaluated for teratogenic potential in rabbits (1 mg/kg/day) is, on a mg/m^2 basis, only about half the MRHD of OM.

OM is transferable to the embryo-fetal compartment. The peak concentration of OM related radioactivity in the rat placenta on the 13th or 18th day of pregnancy (determined in a single dose study with ^{14}C-OM administered at 5 mg/kg) was about 14 or 17%, respectively, of that in the maternal blood. While undetectable in placenta 48 hr after dosing when given on day 13 of gestation, it was still detectable 48 hr after dosing when administered on day 18 of gestation. The concentration of test substance in the amniotic fluid and fetal tissues (especially the digestive tract where it was the highest) increased with time and surpassed the maternal blood concentration (at 24 and 48 hours). Elimination of the radioactivity from the fetus was slower than from maternal tissues. Low concentrations of radioactivity transferred into the milk at a rate slower than into the plasma and transferred out at a rate faster than out of plasma. The milk to plasma exposure ratio for radioactivity (AUC ratio) was about 1.8 : 7.8.

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TABLE 4.4

DOSAGE THRESHOLDS[¶] FOR ADVERSE EFFECTS OF OLMESARTAN MEDOXOMIL IN RAT AND RABBIT REPRODUCTION STUDIES

Report #TR/Review Section #	141-121/3.5.1	140-064/3.5.2	141-086/3.5.3	143-010/3.5.5	143-287/3.5.6	142-147/3.5.4
Species (strain)	Rat (Crj:CD)	Rat (Crj:CD)	Rat (Crj:CD)	Rat (Crj:CD)	Rat (Crj:CD)	Rabbit[a]
Doses (mg/kg/day by gavage)	40, 200, 1000	40, 200, 1000	40, 200, 1000	8, 40, 200	0.3, 1.6	0.3, 1, 3
Days of Drug Administration	See below[b]	GDs 7-17	GDs 7-17	GD 17 - LD 21	GD 17 - LD 21	GD 6-18
Day of Necropsy	GD 20	GD 20	GD 20, LD 22	LD 22	LD 22	GD 28
Maternal Toxicity, F_0						
1. Mortality	>1000	>1000	40-200[f]	>200	>1.6	0.3-1[t]
2. ↓ weight gain	<40[c]	>1000	40-200[g]	40-200[m]	>1.6	>3
3. ↓ food intake	<40[d]	40-200	40-200[h]	8-40[n]	>1.6	0.3-1
4. ↓ corpora lutea	<40[e,*]	>1000	NA	NA	NA	>1[u]
Embryo/Fetal Toxicity, F_1					NA	
1. ↓ survival	>1000	>1000	>1000	NA		>1[v]
2. ↓ fetal weight /pup birth weight	>1000	>1000	40-200[*]	<8		>1
3. Bone variant	>1000	>1000	200-1000[i]	NA		>1
4. Dilatation of the renal pelvis	>1000	>1000	40-200[*]	NA		>1
Neonatal Toxicity, F_1	NA	NA				NA
1. ↓ survival			<40[j]	>200	>1.6	
2. ↓ body weight gain			<40[k,*]	<8[o]	0.3-1.6[r]	
3. Delay in developmental landmarks			8-40[p]	<8[p]	>1.6	
4. Delay in motor coordination			>1000	8-40[q]	>1.6	
5. Dilatation of renal pelvis			>1000	<8[*]	>1.6[s]	
6. Polycystic kidney			>1000	8-40	>1.6	
Adult Reproductive Toxicity, F_1	NA	NA				NA
1.↓ # of implantations			200-1000	>200	>1.6	
2.↓ survival of F_2 pups			40-200[l*]	>200	>1.6	
3.↓ birth weight of F_2 pups			>1000	>200	>1.6	

Footnotes for Table 4.4.

1: When two doses are given, the first is the highest dose at which an effect was not seen and the second is the lowest dose at which an effect was seen.
*: incidence increase with dose
a: Japanese white rabbits
b: 14 days prior to mating with treated males, during mating and until day 7 of gestation.
c: significant ($p < 0.05$) decrease for all dose groups from GD 3 until the day of necropsy.
d: significant ($p < 0.05$) decrease for all dose groups on day 11 of mating and from GD 0 until the day of necropsy.
e: significant ($p < 0.05$) decrease for all dose groups. This effect may not be treatment-related since there are no differences in the number of implantations, post-implantation losses or number of surviving fetuses.
f: among dams assigned to natural delivery, 4 of 13 dams in the high dose group (2 on GD 22, 1 during the labor, and 1 on LD 11) and 1 of 13 dams in the mid dose group (on LD 11) died. Cause of death not given.
g: significant decrease ($p < 0.05$) from GDs 11-20 and LDs 1 to 15.
h: significant decrease ($p < 0.05$) from GDs 9-20.
i: significant decrease ($p < 0.01$) (12% relative to control) in the number of ossified caudal vertebrae in the high dose group.
j: decrease ($p < 0.05$) in the survival rate of pups on postnatal days 1 to 4 in all dose groups.
k: no significant decrease ($p > 0.05$) from LD 4 to 15; significant decrease ($p < 0.05$) from postpartum days 22 to 57 in mid and high dose groups; significant decrease ($p < 0.05$) in low dose group males from postpartum days 43 to 57.
l: significant and dose-dependent decrease ($p < 0.05$) in the number of F_2 pups delivered by F_1 dams from 200 and 1000 mg/kg/day groups.
m: significant reduction ($p < 0.01$) during lactation period, day 4 and 8
n: significantly lower ($p < 0.05$) than control in mid and high dose groups on GD 20 through LD 15 (up to LD 22 in the high dose group)
o: significant reduction ($p < 0.05$) in all dose groups at birth, throughout lactation and up to postnatal day 50 (except for the mid dose group on postnatal day 50)
p: a delay in the mean day of occurrence of separation of ear auricula, eruption of lower incisors, appearance of abdominal hair, separation of eyelids, descent of testes and opening of vagina) in all treated groups. However, the differences from the control group were not statistically significant.
q: motor coordination tests (righting reflex, negative geotaxis) showed no positive reactions or showed prolongation of reaction latency period at the initial testing.
r: significant decrease ($p < 0.05$) compared to control during both pre- and post-weaning days.
s: dilatation of renal pelvis was seen in all dose groups including control. Difference from control not statistically significant.
t: eight of 9 high dose does died between GDs 12 and 17, and 7 of 25 mid dose does died between GDs 14 and 20. Stomach ulcers were observed in all dead animals. Data from the high dose group is excluded from the table.
u: for all practical purposes, 1 mg/kg was considered to be the highest dose studied, see footnote above.
v: no significant effect on fetal survival when analyses limited to surviving dams.

3 pages redacted from this section of the approval package consisted of draft labeling

6. RECOMMENDATIONS

This new drug application for olmesartan medoxomil is approvable with recommended changes in labeling (see pages 219-221).

/S/

G. Jagadeesh, Ph.D.

Cc:
Original NDA 21,286 (Olmesartan medoxomil)
HFD-110
HFD-110/CSO
HFD-110/ Dr. G. Jagadeesh
HFD-124/Dr. J. DeGeorge
HFD-345
Accepted by: /S/ on 10/17/01