SUMMARY OF DRUG WEEK 4 MEAN TOXICOKINETIC PARAMETERS OF L-754,030 IN MALE AND FEMALE RATS FOLLOWING INTRAVENOUS DOSING WITH L-758,298						
	Dose of L-758,298 (mg/kg/day)					
	Males			Females		
Toxicokinetic Parameters	0.25	1	4	0.25	1	4
L-754,030						
C_{max} (µg/ml)	0.100	0.356	1.069	0.127	0.377	1.212
T_{max} (hr)	0.07	0.07	0.07	0.07	0.07	0.07
AUC of the Means (µg•hr/ml)	0.232	0.895	2.737	0.937	2.361	5.621

Study title: **5-Week intravenous toxicity study in rats (TT#97-002-0)**

Study no: TT#97-002-0

Conducting laboratory and location: Merck Research Laboratories, West Point, PA.

Date of study initiation: January 9, 1997

Date of study report: July 29, 1997

GLP compliance: A statement of compliance is included.

QA report: yes (X) no ( )

Drug, lot #, and % purity: L-758, 298. Lot No. 003C009. 98.6% pure.

Formulation/vehicle: Tween-sodium citrate diluent (TSCD), pH 7.5. The composition (per ml) of the vehicle is as follows: Polysorbate 80 (Tween 80)-5.0 mg; D-lactose-10.0 mg; mannitol-10.0 mg; meglumine-1.24 mg; sodium citrate (dihydrate)-5.88 mg; citric acid (anhydrous)-0.38 mg; sodium chloride-9.0 mg.

Methods:

Dosing:

Species/strain: Sprague Dawley rat (Crl:CD®(SD)BR)

#/sex/group or time point (main study): 15/sex/group

Satellite groups used for toxicokinetics or recovery: None

Age: 39 days

Weight: Males: 138 to 189 grams; Females: 114 to 150 grams

Doses in administered units: 2, 5, and 10 mg/kg/day. The basis of dose selection was not mentioned.

Route, form, volume, and infusion rate: Intravenous, solution, 5 ml/kg, and 2 ml/min

The following table presents the study design:

Group No.	Treatment	Dose (mg/kg/day)	Concentration (mg/ml)	Dose Volume (ml/kg)	No. of Males	No. of Females
Control 1	Saline	0	0	5	15	15
Control 2	TSCD	0	0	5	15	15
3	L-758, 298	2	0.4	5	15	15
4	L-758, 298	5	1.0	5	15	15
5	L-758, 298	10	2	5	15	15

Observations and times:

Clinical signs: Daily

Body weights: Weekly

Food consumption: Once or twice weekly by visual inspection

Ophthalmoscopy: Week 4

Hematology: Week 2 and 4

Clinical chemistry: Week 2 and 4

Urinalysis: Week 4

Gross pathology: At necropsy

Organs weighed: At necropsy. The following organs were weighed: heart, spleen, brain, pituitary, kidneys, testes, prostate, thyroid, liver, adrenal, and ovaries.

Histopathology: At necropsy from control-2 and 10 mg/kg/day groups. The following organs or tissues were examined: lung, heart, liver, kidneys, urinary bladder, spleen, thymus, lymph nodes, adrenals, thyroid with parathyroid, pituitary, salivary gland, stomach, small intestine, injection site (tail), pancreas, spinal cord, peripheral nerves, eye with optic nerve and Harder's gland, skin (with mammary gland), bone (including joint), bone marrow, testes and epididymides, prostate, ovaries, uterus, brain, skeletal muscle, large intestine and esophagus.

Toxicokinetics: Plasma samples were taken from 3 rats/sex/timepoint at Week 4.

Results:

1. **Clinical Signs:** Treatment-induced salivation was observed in all groups during week 4.
2. **Mortality:** None.
3. **Body Weight:** The mean initial and final body weights of the Control-2 (vehicle-treated) males were 170 g and 266 g, respectively. The mean initial and final body weights of the Control-2 (vehicle-treated) females were 129 g and 176 g, respectively. There were no treatment-related changes in the body weight in either sex.
4. **Food Consumption:** Food consumption was observed visually and there were no apparent treatment-related changes in food consumption.
5. **Hematology:** No treatment-related changes were observed.
6. **Blood Chemistry:** No treatment-related changes were observed.
7. **Urinalysis:** No treatment-related changes were observed.
8. **Ophthalmology:** No treatment-related changes were observed.
9. **Organ Weights:** Liver weights were increased in a dose-related manner in the females (absolute: 104, 117, and 122% of control at 2, 5, and 10 mg/kg/day, respectively, control = 5.78 g; relative (% body weight): 106, 116, and 122% of control at 2, 5, and 10 mg/kg/day, respectively, control = 3.43%; relative (% brain weight): 103, 116, and 120% of control at 2, 5, and 10 mg/kg/day, respectively, control = 320%) as well as in the males (absolute: 104, 110, and 115% of control at 2, 5, and 10 mg/kg/day, respectively, control = 8.13 g; relative (% body weight): 104, 107, and 113% of control at 2, 5, and 10 mg/kg/day, respectively, control = 3.26%; relative (% brain weight): 103, 108, and 114% of control at 2, 5, and 10 mg/kg/day, respectively, control = 418%). These changes in the liver weight were considered to be due to hepatocellular hypertrophy seen in the females in these groups. However, no such histopathological changes in the liver were observed in the males. The apparent cause of increased liver weight in males was not clear. However, it is to be mentioned here that in a previous i.v. 5-week toxicity study in rats at 0.25, 1, and 4 mg/kg (TT#95-607-0), the females showed 2 to 4-fold higher exposure to active drugs than males. It is possible

that in the present study, the sex-specific histopathological changes in the liver (hepatocellular hypertrophy in females) could be partially attributed to the sex-related difference in exposure to active drug.

10. **Gross Pathology:** No treatment-related changes were observed.

11. **Histopathology:** Hepatocellular hypertrophy was seen in the livers of 5 of 15 females and 6 of 15 females at 5 and 10 mg/kg/day, respectively. No treatment-related changes in the liver were seen in the males. There were no other significant treatment-related changes.

12. **Toxicokinetics:** Blood samples were collected from 3 rats/sex/timepoint at approximately 4 min, 8 min, 1, 2, 5, 7, 10 and 24 hours postdose. The sponsor stated that the results of toxicokinetic analysis would be submitted later.

Five-Week Oral Range-Finding and Toxicokinetic Study in Rats (Report Date/Number #97-060-0).

Testing Laboratory: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486

Date Started: July 10, 1997

Date Completed: March 4, 1998

GLP Compliance: Statements of compliance with GLP Regulations and the Quality Assurance Unit were included.

Animals: Sprague Dawley [Crl:CD®(SD)BR] rats were used in these studies. At the start of treatment, animals were 46 days of old and body weight ranges were 132-213 g for male rats and 111-147 g for female rats.

Drug Batch: L-754,030-004H (Lot #21).

Methods: Rats received L-754,030 by oral gavage at doses of 0 mg/kg/day, 0 mg/kg/day-B.I.D., 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. for 28-29 days. There were 15 rats/sex/group. Control rats received the vehicle, 0.5% aqueous methylcellulose containing 0.02% sodium lauryl sulfate. The dose volume was 5 mL/kg, once or twice per day. For B.I.D. groups, the vehicle or L-754,030 was administered a minimum of 5-6 hr apart. Rats were examined daily for mortality and clinical signs of toxicity. Body weight was measured prior to the

start of treatment, once/week in weeks 1 and 4, and twice/week in weeks 2 and 3. Food consumption was measured twice per week. Ophthalmic examinations were performed during week 3 on control-B.I.D. and 1000 mg/kg/day B.I.D. groups. Blood for determination of hematological and clinical chemistry parameters was collected in week 4. Urine for analysis was collected from 10 rats/sex/group during week 4. Blood for determination of plasma drug levels was collected in week 4 at 2, 4, 6, 8, 10, 16, and 24 hr after the first dose. For B.I.D. groups, the second dose was administered immediately after collection of blood at the 6-hr time point. Four rats/sex/group were bled at 2 and 8 hr, 4 and 10 hr, or 6 and 16 hr and 3 rats/sex/group were bled at 24 hr. Blood samples (2 mL) were also collected from control groups to match treatment groups. L-754,030 was isolated by [redacted]. At the termination of treatment, all animals were sacrificed and a complete gross examination was performed on each animal. Organ weights were measured for the heart, spleen, brain, pituitary, kidneys, testes, prostate, thyroid gland, liver, adrenal glands, and ovaries. Organ weights were expressed as absolute, percent of body weight, and percent of brain weight. A complete microscopic examination for organs and tissues from the control-B.I.D. and 1000 mg/kg/day-B.I.D. groups was performed as follows: lung, heart, liver, kidneys, urinary bladder, spleen, thymus, lymph nodes, adrenal glands, thyroid gland (with parathyroid), pituitary gland, salivary gland, stomach, small intestine, pancreas, spinal cord, peripheral nerve, eye (with optic nerve and Harder's gland), skin (with mammary gland), bone (including joint), bone marrow, testes and epididymides, prostate, ovaries, esophagus, uterus, brain, skeletal muscle, and large intestine. In addition, all grossly observed changes in the liver and thyroid gland from all animals and all grossly changes in the pituitary gland from all male rats were subjected to microscopic examination.

Results:

1. Observed Effects: There were no treatment-related observed effects.

2. Mortality: There were no treatment-related deaths. One male rat in the 1000 mg/kg/ day-B.I.D. died during the blood collection procedure during week 4 at the 16 hr time point.

3. Body Weight and Food Consumption: There were no treatment-related changes of body weight gain or food consumption. Body weights for the male control group during weeks –1 and 4 were 176 and 185 g, respectively. Body weights for the male control-B.I.D. group during weeks –1 and 4 were 177 and 281 g, respectively. Body weight gains for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were 110.9, 111.9, 109.9, and 104.2% of controls, respectively. Body weights for the female control group during weeks –1 and 4 were 134 and 185 g, respectively. Body weights for the female control-B.I.D. group during weeks –1 and 4 were 130 and 184 g, respectively. Body weight gains for the female 250 mg/kg/day, 250 mg/kg/ day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were 99.8, 100.6, 103.2, and 96.3% of controls, respectively.

4. Hematology: There were no treatment-related changes of hematological parameters.

5. Blood Biochemistry and Urinalysis: Serum protein levels for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 103.4, 103.4, 105.2, and 106.9% of controls (Control and Control-B.I.D. values were both 5.8 g/dL), respectively. Serum protein levels for the female 250 mg/kg/ day, 250 mg/kg/day-B.I.D., 500

mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 106.8, 111.7, 108.3, and 108.3% of controls (Control and Control-B.I.D. values were 5.9 and 6.0 g/dL, respectively), respectively. The albumin to globulin ratio for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/ day-B.I.D. groups were decreased to 92.3, 92.3, 84.6, and 84.6% of controls (Control and Control-B.I.D. values were both 1.3), respectively. The albumin to globulin ratio for the female 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were decreased to 86.7, 80, 80, and 80% of controls (Control and Control-B.I.D. values were both 1.5), respectively. Serum calcium levels for the female 250 mg/ kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 103, 105, 102, and 104% of controls (Control and Control-B.I.D. values were both 10 mg/mL), respectively. Cholesterol levels for the male 1000 mg/kg/day-B.I.D. group were increased to 108.6% of the control (58 mg/dL). Cholesterol levels for the female 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 112.5, 178.9, 162, and 171.8% of controls (Control and Control-B.I.D. values were 80 and 71 mg/dL, respectively), respectively. Triglyceride levels for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/ day-B.I.D. groups were decreased to 68.75, 57.7, 57.7, and 52.1% of controls (Control and Control-B.I.D. values were 64 and 71 mg/dL, respectively), respectively. There were no treatment-related changes of urinalysis parameters.

6. Ophthalmic Examination: There were no treatment-related ophthalmic effects.

7. Organ Weights: Treatment-related increases in absolute and relative liver and thyroid gland weights were observed for all male and female treatment groups. Increased liver weight was correlated with hepatocellular hypertrophy. Increased thyroid gland weight was correlated with thyroid follicular cell hyperplasia.

Liver: Absolute liver weights for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 125.2, 141.1, 148, and 148.8% of controls (Control and Control-B.I.D. values were 9.37 and 9.17 g, respectively), respectively. Relative liver weights for the male 250 mg/kg/day, 250 mg/kg/ day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 125.2, 136.2, 142.4, and 149.6% of control (Control and Control-B.I.D. values were 3.41 and 3.37%, respectively), respectively. Absolute liver weights for the female 250 mg/kg/ day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 146.2, 194.4, 170.6, and 187.3% of controls (Control and Control-B.I.D. values were 6.51 and 6.78 g, respectively), respectively. Relative liver weights for the female 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 143.3, 192.7, 173.3, and 181.7% of control (Control and Control-B.I.D. values were 3.55 and 3.71%, respectively), respectively.

Thyroid Gland: Absolute thyroid gland weights for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 112.7, 135.2, 139.3 and 133.1% of controls (Control and Control-B.I.D. values were 0.0158 and 0.0145 g, respectively), respectively. Relative thyroid gland weights for the male 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/ day-B.I.D. groups were increased to 114, 129.6, 131.5, and 133.3% of control (Control and Control-B.I.D. values were 0.0057 and 0.0054%, respectively), respectively. Absolute thyroid gland weights for the female 250 mg/kg/day, 250

mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 122.8, 125.8, 119.6, and 135.8% of controls (Control and Control-B.I.D. values were 0.0123 and 0.0120 g, respectively), respectively. Relative liver weights for the female 250 mg/kg/day, 250 mg/kg/ day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. groups were increased to 119, 124.2, 119.7, and 131.8% of control (Control and Control-B.I.D. values were 0.0068 and 0.0066%, respectively), respectively.

8. Gross Pathology: Necropsy examination found that the livers of several female rats in the 1000 mg/kg/day B.I.D. group were enlarged and/or red/yellow in color. The sponsor did not provide a summary table or individual line listings for gross pathological changes.

9. Histopathology: Target organs of toxicity were the liver, thyroid gland, and pituitary gland. For the liver, very slight to slight hepatocellular hypertrophy was observed in all male and female treatment groups. Hepatocyte hypertrophy most likely represents an induction of cytochrome P-450 enzymes and has no toxicological significance. Very slight to slight diffuse vacuolation was observed with an increased incidence for male and female rats that received doses of 250, 500, and 1000 mg/kg/day-B.I.D. For the thyroid gland, very slight to slight thyroid follicular cell hyperplasia was observed was observed in all male and female treatment groups. Follicular cell hyperplasia of the thyroid gland is most likely due to increased catabolism of thyroxine (T_4) and triiodothyronine (T_3) by the liver. For the pituitary gland, a very slight to slight dose-dependent vacuolation of individual cells in the pars distalis was observed for all male treatment groups. This change was characterized by enlargement of individual pituitary cells due to formation of large cytoplasmic vacuoles and occasional protein droplets. This change may represent a degeneration or exhaustion of Thyroid stimulating hormone-producing pituitary cells secondary to hepatic enzyme induction and increased catabolism of T_3 and T_4.

Histopathological changes for rats that received L-754,030 by oral gavage at doses of 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. for 28 or 29 days (n = 15/group).

Organ/Tissue	0		0-B.I.D.		250		250-B.I.D.		500-B.I.D.		1000-B.I.D.	
	F	M	F	M	F	M	F	M	F	M	F	M
Liver -hepatocyte hypertrophy -diffuse vacuolation	 0 1	 0 0	 0 3	 0 0	 14 2	 8 1	 15 5	 15 3	 15 8	 15 4	 15 8	 14 3
Thyroid gland -follicular cell, diffuse hyperplasia	0	0	0	0	8	4	15	9	14	11	10	10
Pituitary gland -vacuolation	-	0	0	0	-	1	-	5	-	8	-	10

10. Plasma Drug Levels: A plateau in plasma C_{max} and $AUC_{0\text{-}24hr}$ values for L-754,030 was evident in male and female treatment groups in week 4 at doses ≥250 mg/kg/day B.I.D. Plasma C_{max} and $AUC_{0\text{-}24hr}$ values for L-754,030 in female rats were significantly higher than in male rats.

Toxicokinetic parameters for plasma L-754,030 levels in weeks for rats that received L-754,030 by oral gavage at doses of 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/ day-B.I.D., and 1000 mg/kg/day-B.I.D.

Dose,	C_{max}, µg/mL		T_{max}, hr		AUC_{0-24hr}, µg*hr/mL	
mg/kg/day	Male	Female	Male	Female	Male	Female
250	0.233	0.819	2	24	3.21	13.9
250-B.I.D.	0.519	2.59	16	24	7.19	33.2
500-B.I.D.	0.444	2.27	16	24	7.22	34.8
1000-B.I.D.	0.722	1.74	16	16	9.37	32.1

Rats received L-754,030 by oral gavage at doses of 0 mg/kg/day, 0 mg/kg/day-B.I.D., 250 mg/kg/day, 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. for 28-29 days. A no effect dose was not established. Target organs of toxicity were the liver, thyroid gland, and pituitary gland. For the liver, very slight to slight hepatocellular hypertrophy was in all male and female treatment groups. Hepatocyte hypertrophy most likely represents an induction of cytochrome P-450 enzymes and has no toxicological significance. Very slight to slight diffuse vacuolation was observed with an increased for male and female rats that received doses of 250 mg/kg/day-B.I.D., 500 mg/kg/day-B.I.D., and 1000 mg/kg/day-B.I.D. For the thyroid gland, very slight to slight thyroid follicular cell hyperplasia was observed for all male and female treatment groups. Follicular cell hyperplasia of the thyroid gland is most likely due to increased catabolism of thyroxine (T_4) and triiodothyronine (T_3) by the liver. For the pituitary gland, a very slight to slight dose-dependent vacuolation of individual cells in the pars distalis was observed for all male treatment groups. This change was characterized by enlargement of individual pituitary cells due to formation of large cytoplasmic vacuoles and occasional protein droplets. This change may represent a degeneration or exhaustion of Thyroid stimulating hormone-producing pituitary cells secondary to hepatic enzyme induction and increased catabolism of T_3 and T_4. A plateau in plasma C_{max} and AUC_{0-24hr} values for L-754,030 was evident in male and female treatment groups in week 4 at doses ≥250 mg/kg/day B.I.D. Plasma C_{max} and AUC_{0-24hr} values for L-754,030 in female rats were significantly higher than in male rats.

Study Title: MK-0869: Five-Week Oral Toxicity Study in Rats.

Report No: TT 00-001-0,-1
Conducting laboratory and location: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486

Date Started: January 6, 2000

Date Completed: August 8, 2000
GLP compliance: A statement of compliance with GLP regulations was included.
QA- Report Yes (X) No ( )
Methods: The toxicology and toxicokinetic profiles of two MK-0869 formulations of different particle size, were assessed in Sprague-Dawley rats during a 5-week treatment period (28 to 29 days). MK-0869 Formulation M (average particle size, —) was administered to 16 rats/sex/group at oral doses of 0 and 125 mg/kg B.I.D. (total daily doses of 0 and 250 mg/kg/day, respectively). MK-0869 Formulation NB (average particle size, —) was administered to 16 rats/sex/group at oral doses of 0, 5, 125, 250, 500, and 750 mg/kg B.I.D. (total daily doses of 0, 10, 250, 500, 1000, and 1500 mg/kg, respectively). The first and second daily doses were administered a minimum of 4 hr apart for vehicle-control groups and a minimum of 6 hr apart for all drug-treated groups. A total of 56 to 58 doses were administered.
Dosing:

- **species/strain:** Sprague-Dawley rats [Crl:CD®(SD)IGS BR] were obtained from
- **#/sex/group or time point:** 16 rats/sex/group
- **age:** Male and female rats were 42 and 38 days old, respectively, at the start of treatment.
- **weight:** Body weight ranges were 131 to 200 g for male rats and 93 to 135 g for female rats at the start of treatment.
- **satellite groups used for toxicokinetics or recovery:** None.
- **dosage groups in administered units:** MK-0869 Formulation M (average particle size, — was administered at doses of 0 and 125 mg/kg B.I.D. (total daily doses of 0 and 250 mg/kg/day, respectively). Formulation NB (average particle size, —) was administered at doses of 0, 5, 125, 250, 500, and 750 mg/kg B.I.D. (total daily doses of 0, 10, 250, 500, 1000, and 1500 mg/kg, respectively). The first and second daily doses were administered a minimum of 4 hr apart for vehicle-control groups and a minimum of 6 hr apart for all drug-treated groups.
- **route, form, volume, and infusion rate:** Vehicle or drug suspension was administered by oral gavage using a dose volume of 5 mL/kg.

Drug, lot#, radiolabel, and % purity: MK-0869 Formulation M, lot number L-754030-004H031, had an average particle size of —µm with 99.5% purity. MK-0869, Formulation NB (MK-0869 blended coated — beads), batch #X0869OPP015C001 (also known as L-754030-016S001) with an average particle size of — was obtained by blending 3 batches of MK-0869 coated beads with purity ranging from 99.6 to 100.0%.
Formulation/vehicle: The vehicle for MK-0869 Formulation M was 5% methylcellulose and 0.02% sodium lauryl sulfate in deionized water. The vehicle for MK-0869 Formulation NB was 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water.
Observations and times:

- **Clinical signs:** Animals were monitored daily for clinical signs of toxicity and mortality before and after dosing.
- **Body weights:** Body weights were measured prior to the start of treatment, once per week during weeks 1 and 2, and 2 per week thereafter.

- **Food consumption:** Cages were examined twice per week at 24 hr after dosing to determine if any food was remaining.
- **Ophthalmoscopy:** Ophthalmic examinations were conducted in control and high dose groups during week 4.
- **EKG:** Not performed.
- **Hematology:** Blood for determination of hematology parameters was collected during weeks 2 and 4.
- **Clinical chemistry:** Blood for determination of clinical chemistry parameters was collected during weeks 2 and 4.
- **Urinalysis:** Urine samples for analysis were collected from approximately 10 rats/sex/group during week 4.
- **Gross pathology:** Rats were sacrificed at the termination of the study and submitted to complete necropsy.
- **Organs weighed:** Absolute and relative organ weights were determined for the adrenal glands, brain, heart, ovaries, kidneys, liver, pituitary gland, prostate gland, spleen, testes, and thyroid gland.
- **Histopathology:** Tissue sections from the vehicle-control groups, Formulation M high dose group, Formulation NB high dose, and deaths or moribund sacrifices during the treatment period were prepared, stained with hematoxylin and eosin, and submitted to microscopic examination. Tissues with gross pathological changes from all animals were examined at the discretion of the pathologist. Sections of liver and thyroid gland were examined from all animals. Tissues examined were as follows: salivary gland (submandibular/sublingual), esophagus, stomach (glandular and nonglandular portions), small intestines (duodenum, jejunum, ileum), large intestine (colon), liver, pancreas, adrenal glands, parathyroid (when present in thyroidal sections), pituitary gland, thyroid gland, kidneys, skin (from mammary region), mammary gland (when present in skin section), lung, heart, spleen, lymph nodes (cervical and mesenteric), thymus, bone marrow, bone (femur, including femorotibial joint), skeletal muscle, brain (including cerebral cortex and subcortical white matter, basal ganglia, cerebellum, and pons), urinary bladder, ovaries, uterus, testes and epididymides, prostate, spinal cord (cervical) peripheral nerve (sciatic), eye (with optic nerve), and Harder's gland.
- **Toxicokinetics:** Blood for determination of plasma drug levels was collected during week 4 at 2, 4, 6, 8, 10, 16, and 24 hr after dosing. Four rats/sex/group were used for each time point. Levels of MK-0869 were quantified by [redacted]. The lower limit of quantitation was [redacted] µg/mL.
- **Other:** None.

Results:

- **Clinical signs:** There were no treatment-related clinical signs of toxicity.

- **Mortality:** There was no treatment-related mortality. Two animals died during the treatment period due to a dosing accident or from anesthesia associated with blood collection. Animal #00-0165F in the Formulation NB 250 mg/kg B.I.D. group was found dead during week 4. Death was attributed to a dosing accident. Animal #00-0237F in the

Formulation NB 750 mg/kg B.I.D. group was found dead during week 4. Death was attributed to an anesthesia accident associated with blood collection

- **Body weights:** There were no treatment-related effects on body weight gains with MK-0869 Formulation M or NB. Body weights for male controls, which received the vehicle for Formulation M, at weeks -1 and 4 were 167 and 278 g, respectively. Body weights for female controls, which received vehicle for Formulation M, at weeks -1 and 4 were 118 and 171 g, respectively. Body weight gains for male and female rats that received MK-0869 Formulation M at 125 mg/kg B.I.D. were 96 and 115% of the control, respectively. Body weights for male controls, which received vehicle for Formulation NB, at weeks -1 and 4 were 172 and 291 g, respectively. Body weight gains for male rats that received MK-0869 Formulation NB at doses of 5, 125, 250, 500, and 750 mg/kg B.I.D. were 103.2, 100, 105.4, 103.75, and 103.1% of the control, respectively. Body weights for female controls, which received vehicle for Formulation NB, at weeks -1 and 4 were 120 and 179 g, respectively. Body weight gains for female rats that received MK-0869 Formulation NB at doses of 5, 125, 250, 500, and 750 mg/kg B.I.D. were 100.8, 106.9, 110.3, 102.5, and 107.8% of the control, respectively.

- **Food consumption:** There were no treatment-related changes of food consumption.

- **Ophthalmoscopy:** Ophthalmic examinations of vehicle-control groups, the high dose MK-0869 Formulation M group, and the high dose MK-0869 Formulation NB group apparently revealed no treatment-related changes; however, no data was provided for independent analysis.

- **Hematology:** Decreased platelet counts were observed for male treatment groups at week 2 that received MK-0869 Formulation NB and female treatment groups at weeks 2 and 4 that received MK-0869 Formulation NB.

MK-0869 Formulation NB, Week 2: Platelet counts for male rats that received MK-0869 Formulation NB at 125, 250, 500, and 750 mg/kg B.I.D. were increased to 122.2, 121.6, 124.8, and 124.8% of the control (1540 x 10^3/mm^3), respectively. Platelet counts for female rats that received MK-0869 Formulation NB at 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 112.4, 135.7, 147.7, 151.3, and 140.1% of the control (1311 x 10^3/mm^3), respectively.

MK-0869 Formulation NB, Week 4: Platelet counts for female rats that received MK-0869 Formulation NB at 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 114.2, 120.5, 124, 122.7, and 116.0% of the control (1222 x 10^3/mm^3), respectively.

- **Clinical chemistry:** Clinical chemistry changes observed for rats that received MK-0869 Formulation M or NB included alterations of total protein levels, A/G ratios, cholesterol levels, and triglyceride levels, which may be correlated with histopathological changes observed in the liver.

MK-0869 Formulation M, Week 2: Total protein levels for male rats at 125 mg/kg B.I.D. were increased to 105.7% of the control (5.3 g/dL). A/G ratios for male and female rats that received 125 mg/kg B.I.D. were decreased to 80 and 87.5% of control values (1.6 and 1.5), respectively. Cholesterol levels for female rats at 125 mg/kg B.I.D. were increased to 116.4% of the control (67 mg/dL), respectively. Triglyceride levels for male rats that received 125 mg/kg B.I.D. were decreased to 82.8% of the control (64 mg/dL).

MK-0869 Formulation M, Week 4: Total protein levels for male rats at 125 mg/kg B.I.D. were increased to 105.45% of the control (5.5 g/dL). Total protein levels for female rats that received 125 mg/kg B.I.D. were increased to 110.5% of the control (5.7 g/mL). A/G ratios for male and female rats that received 125 mg/kg B.I.D. were decreased to 85.7 and 75% of control values (1.4 and 1.6), respectively. Cholesterol levels for female rats at 125 mg/kg B.I.D. were increased to 135.3% of the control (68 mg/dL), respectively. Triglyceride levels for male rats that received 125 mg/kg B.I.D. were decreased to 62.2% of the control (90 mg/dL).

MK-869 Formulation NB, Week 2: Glucose levels for female rats that received 250, 500, and 750 mg/kg B.I.D. were decreased to 84.1-90.85% of the control (164 mg/dL). Glucose levels for male rats that received 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 89.1-91.5% of the control (165 mg/dL). Total protein levels for male rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 105.5-109.3% of the control (5.4 g/dL). A/G ratios for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 84.1-90.85% of the control (1.6). A/G ratios for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 80-86.7% of the control (1.5). Cholesterol levels for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 111.5-124.4% of the control (78 mg/dL). Cholesterol levels for male rats that received 750 mg/kg B.I.D. were increased to 112.1% of the control (58 mg/dL). Potassium levels for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 106.25-110.4% of the control (4.8 mEq/L). Triglyceride levels for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 85.9, 56.25, 73.4, 54.7, and 64.1% of the control (64 mg/dL), respectively.

MK-0869 Formulation NB, Week 4: Glucose levels for male rats that received 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 82-87.4% of the control (183 mg/dL). Total protein levels for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 110.3-113.8% of the control (5.8 g/mL). Total protein levels for male rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 105.45-110.9% of the control (5.5 g/dL). A/G ratios for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 75% of the control (1.6). A/G ratios for male rats that received 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 85.7% of the control (1.4). Cholesterol levels for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 152-169% of the control (77 mg/dL). Cholesterol levels for male rats that received 750 mg/kg B.I.D. were increased to 117.9% of the control (56 mg/dL). Potassium levels for female rats that received 125, 250, 500, and 750 mg/kg B.I.D. were increased to 104.2-106.25% of the control (4.8 mEq/L). Triglyceride levels for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were decreased to 82.9, 43.9, 63.4, 52.4, and 59.8% of the control (82 mg/dL), respectively.

- **Urinalysis:** There were no treatment-related changes of urinalysis parameters.

- **Organ Weights:** Treatment-related changes of absolute and relative liver and thyroid gland weights were observed with MK-0869 Formulations M and NB.

MK-0869 Formulation M: Absolute and relative liver weights for male rats that received 125 mg/kg B.I.D. were increased to 144.1 and 138.2% of control values (9.87 g and 3.61% B.W.), respectively. Absolute and relative liver weights for female rats that received 125 mg/kg B.I.D. were increased to 177 and 170.6% of control values (6.35 g and 3.74% B.W.), respectively. Absolute and relative thyroid gland weights for male rats that received 125 mg/kg B.I.D. were increased to 136 and 130% of control values (0.0164 g and 0.060% B.W.), respectively. Absolute and relative thyroid gland weights for female rats that received 125 mg/kg B.I.D. were increased to 128.7 and 123.7% of control values (0.0129 g and 0.0076% B.W.), respectively.

MK-0869 Formulation NB: Absolute liver weights for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 109.7, 146.4, 150.7, 150.7, and 156.3% of the control (10.47 g), respectively. Relative liver weights for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 111.5, 148.4, 154.4, 158.5, and 156.3% of the control (3.66% B.W.), respectively. Absolute liver weights for female rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 121.1, 186.1, 206.2, 218.9, and 202.3% of the control (6.62 g), respectively. Relative liver weights for female rats that received 5, 125, 250, 500, and 750 mg/kg were increased to 122.25, 188.2, 206.2, 214.5, and 203.2% of the control (3.73% BW), respectively. Absolute thyroid gland weights for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 108.5, 129.5, 133, 129.5, and 128.4% of the control (0.0176 g), respectively. Relative thyroid gland weights for male rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 111.5, 132.8, 137.7, 137.7, and 129.5% of the control (0.0061% B.W.), respectively. Absolute thyroid gland weights for female rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 108.9, 126.7, 127.4, 136.3, and 141.5% of the control (0.0135 g), respectively. Relative thyroid gland weights for female rats that received 5, 125, 250, 500, and 750 mg/kg B.I.D. were increased to 110.5, 128.9, 128.9, 134.2, and 142.1% of the control (0.0076% B.W.), respectively.

- **Gross Pathology:** Treatment-related liver enlargement, often with prominent lobular architecture, was observed for all MK-0869 Formulation M or NB groups. Treatment-related thyroid gland enlargement was observed for all MK-0869 Formulation M or NB groups.

- **Histopathology:** Histopathological changes were observed in the liver and thyroid gland. Treatment-related hypertrophy of hepatocytes and diffuse vacuolation of hepatocytes were observed in the liver for all MK-0869 Formulation M or NB groups. Treatment-related diffuse follicular cell hyperplasia was observed in the thyroid gland for all MK-0869 Formulation M or NB groups. Benign parafollicular cell adenomas were observed for two animals, one that received MK-0869 Formulation M at 125 mg/kg B.I.D. and one that received MK-0869 Formulation NB at 250 mg/kg B.I.D.

Histopathological changes for rats that received MK-0869 Formulation M at oral doses of 0 or 125 mg/kg B.I.D. for 28 or 29 days.

Tissue	0 mg/kg B.I.D.		125 mg/kg B.I.D.	
	F	M	F	M
Liver				
-hepatocyte hypertrophy	0	0	16	14
-hepatocyte, diffuse vacuolation	0	0	8	2
Thyroid gland				
-parafollicular cell, benign adenoma	0	0	0	1
-follicular cell, diffuse hyperplasia	0	2	11	12

Histopathological changes for rats that received MK-0869 Formulation NB at oral doses of 0. 5, 125, 250, 500, or 750 mg/kg B.I.D. for 28 or 29 days.

Tissue	0		5		125		250		500		750	
	F	M	F	M	F	M	F	M	F	M	F	M
Liver												
-hepatocyte hypertrophy	0	0	9	4	16	16	15	16	16	16	16	16
-hepatocyte,diffuse vacuolation	0	0	0	0	9	4	6	2	3	5	4	3
Thyroid gland												
-parafollicular cell, benign adenoma	0	0	0	0	0	0	1	0	0	0	0	0
-follicular cell,diffuse hyperplasia	0	0	2	6	14	14	12	14	15	16	12	16

- **Toxicokinetics:** Toxicokinetic analysis was restricted to the parent compound, MK-0869. Plasma AUC values for female treatment groups that received MK-0869 Formulation M or NB were significantly higher than AUC values for corresponding male treatment groups. Plasma AUC values for male and female rats that received MK-0869 Formulation M at 125 mg/kg B.I.D. were similar to AUC values for male and female rats that received MK-0869 Formulation NB at 125 mg/kg B.I.D. For male and female rats that received MK-0869 Formulation N at doses of 5 to 750 mg/kg B.I.D., C_{max} and AUC increased in a manner that was significantly less than proportional to dose. For male rats that received MK-0869 Formulation NB at doses from 125 to 750 mg/kg B.I.D., AUC values increased by only 1.6-fold over this 6-fold increase in dose. For female rats that received MK-0869 Formulation NB at doses from 125 to 750 mg/kg B.I.D., AUC values increased by only 1.4-fold over this 6-fold increase in dose. A plateau in AUC values for male rats that received MK-0869 Formulation NB was evident at approximately 500 mg/kg B.I.D. A plateau in AUC values for female rats that received MK-0869 Formulation NB was evident at approximately 250 mg/kg B.I.D.

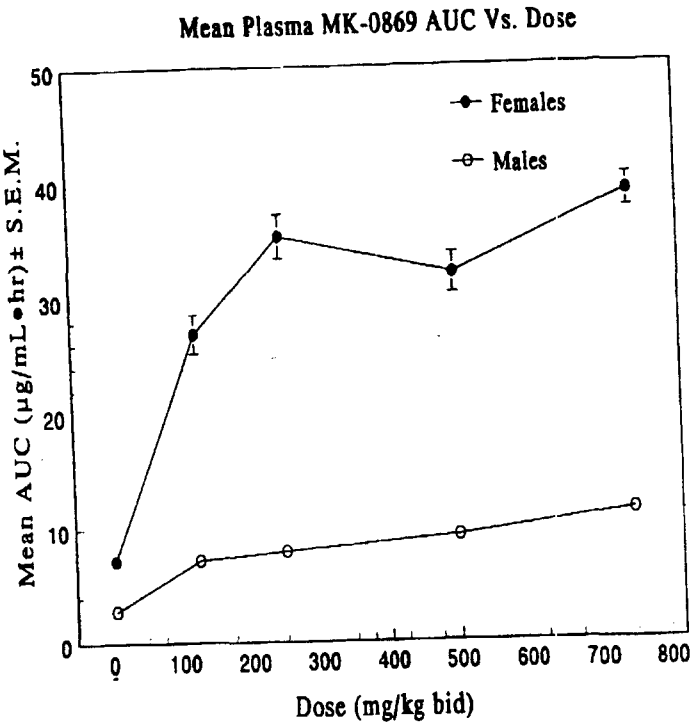
Plasma toxicokinetic parameters of MK-0869 in male and female rats during week 4 following B.I.D. dosing with MK-0869 in Formulation NB

Dose, mg/kg B.I.D.	$AUC_{0\text{-}24hr}$, µg·hr/mL		C_{max}, µg/mL		T_{max}, hr	
	Male	Female	Male	Female	Male	Female
5	2.82	7.20	0.340	0.724	2.0	2.0
125	7.19	26.9	0.485	1.66	8.0	8.0
250	7.91	35.3	0.487	1.91	8.0	24
500	9.21	32.0	0.612	1.63	8.0	8.0
750	11.3	38.9	0.660	2.32	10	10

Plasma toxicokinetic parameters of MK-0869 in male and female rats during week 4 following B.I.D. dosing with MK-0869 in Formulation M.

Dose, mg/kg B.I.D.	$AUC_{0\text{-}24hr}$, µg·hr/mL		C_{max}, µg/mL		T_{max}, hr	
	Male	Female	Male	Female	Male	Female
125	5.28	19.0	0.345	1.32	24	2.0

Dose and Sex Comparison of Drug Week 4 Mean Plasma MK-0869 AUC Values in Rats Treated With B.I.D. Doses of MK-0869 NB Formulation



Key Study Findings: The toxicology and toxicokinetic profiles of two MK-0869 formulations of different particle size, were assessed in Sprague-Dawley rats during a 5-week treatment period (28 to 29 days). MK-0869 Formulation M (average particle size, ~~~) was administered to 16 rats/sex/group at oral doses of 0 and 125 mg/kg B.I.D. (total daily doses of 0 and 250 mg/kg/day, respectively). MK-0869 Formulation NB (average particle size, ~~~ was administered to 16 rats/sex/group at oral doses of 0, 5, 125, 250, 500, and 750 mg/kg B.I.D. (total daily doses of 0, 10, 250, 500, 1000, and 1500 mg/kg/day, respectively).

The first and second daily doses were administered a minimum of 4 hr apart for vehicle-control groups and a minimum of 6 hr apart for all drug-treated groups. A total of 56 to 58 doses were administered. A no effect dose was not observed with Formulation M or NB. Histopathological changes were observed in the liver and thyroid gland. Treatment-related hypertrophy of hepatocytes and diffuse vacuolation of hepatocytes were observed in the liver for all MK-0869 Formulation M or NB groups. Treatment-related diffuse follicular cell hyperplasia was observed in the thyroid gland for all MK-0869 Formulation M or NB groups. Benign parafollicular cell adenomas were observed for two animals, one that received MK-0869 Formulation M at 125 mg/kg B.I.D. and one that received MK-0869 Formulation NB at 250 mg/kg B.I.D. Toxicokinetic analysis was restricted to the parent compound, MK-0869. Plasma AUC values for female treatment groups that received MK-0869 Formulation M or NB were significantly higher than AUC values for corresponding male treatment groups. Plasma AUC values for male and female rats that received MK-0869 Formulation M at 125 mg/kg B.I.D. were similar to AUC values for male and female rats that received MK-0869 Formulation NB at 125 mg/kg B.I.D. For male and female rats that received MK-0869 Formulation N at doses of 5 to 750 mg/kg B.I.D., C_{max} and AUC increased in a significantly less than proportional manner to dose. For male rats that received MK-0869 Formulation NB at doses from 125 to 750 mg/kg B.I.D., AUC values increased by only 1.6-fold over this 6-fold increase in dose. For female rats that received MK-0869 Formulation NB at doses from 125 to 750 mg/kg B.I.D., AUC values increased by only 1.4-fold over this 6-fold increase in dose. A plateau in AUC values for male rats that received MK-0869 Formulation NB was evident at approximately 500 mg/kg B.I.D. A plateau in AUC values for female rats that received MK-0869 Formulation NB was evident at approximately 250 mg/kg B.I.D.

Fourteen-Week Oral Range-Finding Study in Rats (Report Date/Number TT #97-117-0).

Testing Laboratory: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486

Date Started: November 3, 1997

Date Completed: July 8, 1998

GLP Compliance: Statements of compliance with GLP Regulations and the Quality Assurance Unit were included.

Animals: Sprague Dawley [Crl:CD®(SD)BR] rats were used in these studies. At the start of treatment, animals were 35 days of old and body weight ranges were 98-150 g for male rats and 101-124 g for female rats.

Drug Batch: L-754,030-004H026

Methods: Rats received L-754,030 by oral gavage at doses of 0, 5, 25, 125, and 250 mg/ kg/day-B.I.D. for 14 weeks. Male rats received a total of 182 doses and female rats received a total of 184 doses. There were 15 rats/sex/group. Control rats received the vehicle, 0.5% aqueous methylcellulose containing 0.02% sodium lauryl sulfate. The dose volume was 5 mL/kg, twice per day. For B.I.D. groups, the vehicle or L-754,030 was administered a minimum of 5-6 hr apart. Animals were fed between the first and second doses. The initiation of feeding was 2-3 hr after the last drug treatment group received its first daily dose and 3-4 hr before administration of the second daily dose. From week 7 to study termination, the initiation of feeding was 2-2.5 hr after the last drug treatment group received their first daily dose and 3-4.5 hr before administration of the second daily dose. Rats were examined daily for mortality and clinical signs of toxicity. Body weight was measured prior to the start of treatment, and once or twice/week, thereafter. Food consumption was estimated twice per week by visual inspection. Ophthalmic examinations were performed during weeks 6 and 12 on control-B.I.D. and 250 mg/kg/day B.I.D. groups. Blood for determination of hematological and clinical chemistry parameters was collected at weeks 5 and 11. Urine for analysis was collected from 10 rats/sex/group during week 11. Blood for determination of plasma drug levels was collected in week 13 at 2, 4, 6, 8, 10, 16, and 24 hr after the first dose. Three to four rats/sex/group were bled at each time point. Blood samples (3 mL) were also collected from control groups to match treatment groups. L-754,030 was isolated by [redacted] and quantified by [redacted] At the termination of treatment, all animals were sacrificed and a complete gross examination was performed on each animal. Organ weights were measured for the heart, spleen, brain, pituitary, kidneys, testes, prostate, thyroid gland, liver, adrenal glands, and ovaries. Organ weights were expressed as absolute, percent of body weight, and percent of brain weight. A complete microscopic examination for organs and tissues from the control-B.I.D. and 250 mg/kg/day-B.I.D. groups was performed as follows: lung, heart, liver, kidneys, urinary bladder, spleen, thymus, lymph nodes, adrenal glands, thyroid gland (with parathyroid), pituitary gland, salivary gland, stomach, small intestine, pancreas, spinal cord, peripheral nerve, eye (with optic nerve and Harder's gland), skin (with mammary gland), bone (including joint), bone marrow, testes and epididymides, prostate, ovaries, esophagus, uterus, brain, skeletal muscle, and large intestine. In addition, all grossly observed changes in the liver, thyroid gland, and pituitary gland from all animals were subjected to microscopic examination.

Results:

1. Observed Effects: There were no treatment-related observed effects.

2. Mortality: There were no treatment-related deaths.

3. Body Weight and Food Consumption: There were no treatment-related changes of body weight gain or food consumption. Body weights for the male control-B.I.D. group at weeks -1 and 13 were 128 and 388 g, respectively. Body weight gains for the male 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were 100.4, 100.8, 98.14, and 101.2% of the control, respectively. Body weights for the female control-B.I.D. group at weeks -1 and 13 were 111 and 232 g, respectively. Body weight gains for the male 5, 25, 125, and 250 mg/ kg/day-B.I.D. groups were 103.5, 102.5, 90.2, and 100.9% of the control, respectively.

4. Hematology: A number of small, statistically significant changes were observed in hematological parameters (i.e., hemoglobin levels, hematocrit, mean corpuscular volume) for male and female

treatment groups; however, changes were generally <5-10% and appeared to have little or no biological significance.

5. Blood Biochemistry and Urinalysis:

Week 5: Protein levels for the female 125 and 250 mg/kg/day-B.I.D. groups were both increased to 108.3% of the control (6.0 g/dL). Protein levels for the male 125 and 250 mg/kg/day-B.I.D. groups were increased to 106.8 and 105.1% of the control (5.9 g/dL), respectively. The albumin to globulin ratio for the female 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were decreased to 92.9, 85.7, 78.6, and 85.7% of the control (1.4), respectively. The albumin to globulin ratio for the male 250 mg/kg/day-B.I.D. group was increased to 92.3% of the control (1.3). Alkaline phosphatase activity for the female 125 and 250 mg/kg/day-B.I.D. groups were decreased to 77.7 and 77.0% of the control (148 U/L), respectively. Cholesterol levels for the female 125 and 250 mg/kg/day-B.I.D. groups were increased to 123.1 and 128.6% of the control (91 mg/dL), respectively. Triglyceride levels for the female 125 and 250 mg/kg/day-B.I.D. groups were decreased to 84.4 and. 75% of the control (64 mg/dL), respectively. Triglyceride levels for the male 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were decreased to 85.5, 82.3, 51.6, and 45.2% of the control (62 mg/dL), respectively.

Week 11: Protein levels for the female 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 103.2, 106.3, 117.5, and 114.3% of the control (6.3 g/dL), respectively. Protein levels for the male 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 103.3, 106.6, and 108.2% of the control (6.1 g/dL), respectively. Albumin levels for the female 125 and 250 mg/kg/day-B.I.D. groups were increased to 108.3 and 105.6 of the control (3.6 g/dL), respectively. The albumin to globulin ratio for the female 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were decreased to 92.3, 92.3, 84.6, and 84.6% of the control (1.3), respectively. The albumin to globulin ratio for the male 5, 25, 125, and 250 mg/kg/ day-B.I.D. groups were decreased to 91.7, 91.7, 83.3, and 83.3% of the control (1.2), respectively. Alkaline phosphatase activity for the female 125 and 250 mg/kg/day-B.I.D. groups were decreased to 67.5 and 71.1% of the control (83 U/L), respectively. Calcium levels for the female 250 mg/kg/day-B.I.D. group were increased to 105.2% of the control (9.7 mg/dL). Cholesterol levels for the female 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 121.95, 145.1, and 158.5% of the control (82 mg/dL), respectively. Triglyceride levels for the female 125 and 250 mg/kg/day-B.I.D. groups were decreased to 88.9 and 84.1% of the control (63 mg/dL), respectively. Triglyceride levels for the male 5, 25, 125, and 250 mg/kg/day groups were decreased to 34.8, 63.3, 44.3, and 43% of the control (79 mg/dL), respectively. There were no treatment-related changes of urinalysis parameters.

6. Ophthalmic Examination: There were no treatment-related ophthalmic effects.

7. Organ Weights: Treatment-related increases in absolute and relative liver and thyroid gland weights were observed for all male and female treatment groups. Increased liver weight was correlated with hepatocellular hypertrophy. Increased thyroid gland weight was correlated with thyroid follicular cell hyperplasia.

Liver: Absolute liver weight for the male 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 111.3, 126.8, 135.4, and 149.9% of the control (10.68 g), respectively. Relative liver weight for the male 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 109.7, 124.9, 132.1, and 145% of the control (2.89%), respectively. Absolute liver weight for the female 5, 25, 125,

and 250 mg/kg/day-B.I.D. groups were increased to 122.8, 148, 179.6, and 180.4% of the control (6.79 g), respectively. Relative liver weight for the female 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 121.9, 145.1, 179.1, and 180.7% of the control (3.06%), respectively.

Thyroid Gland: Absolute thyroid gland weight for the male 5, 25, 125, and 250 mg/ kg/day-B.I.D. groups were increased to 127.7, 129.7, 129, and 150% of the control (0.0148 g), respectively. Relative thyroid gland weight for the male 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 125, 127.5, 127.5, and 145% of the control (0.004%), respectively. Absolute thyroid gland weight for the female 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 113.7, 119.8, 118.3, and 138.2% of the control (0.0131 g), respectively. Relative thyroid gland weight for the female 5, 25, 125, and 250 mg/kg/day-B.I.D. groups were increased to 113.6, 118.6, 118.6, and 139% of the control (0.0059%), respectively.

8. Gross Pathology: The sponsor did not provide a summary table or individual line listings for gross pathological changes.

9. Histopathology: Target organs of toxicity were the liver, thyroid gland, and pituitary gland. For the liver, very slight to slight hepatocellular hypertrophy was observed in all male and female treatment groups. Hepatocyte hypertrophy most likely represents an induction of cytochrome P-450 enzymes and has no toxicological significance. Very slight to slight diffuse hepatocellular vacuolation was observed with an increased incidence for male rats that received a dose of 250 mg/kg/day-B.I.D. For the thyroid gland, very slight to slight diffuse thyroid follicular cell hyperplasia was observed for all male and female treatment groups. Follicular cell hyperplasia of the thyroid gland is most likely due to increased catabolism of thyroxine (T_4) and triiodothyronine (T_3) by the liver. For the pituitary gland, a very slight to slight vacuolation of individual cells in the pars distalis was observed for male rats that received doses of 125 and 250 mg/kg/day-B.I.D. This change was characterized by enlargement of individual pituitary cells due to formation of large cytoplasmic vacuoles and occasional protein droplets. This change may represent a degeneration or exhaustion of Thyroid stimulating hormone-producing pituitary cells secondary to hepatic enzyme induction and increased catabolism of T_3 and T_4. For the adrenal gland, diffuse vacuolation was observed in the cortex.

Histopathological changes for rats that received L-754,030 by oral gavage at doses of 0, 5, 25, 125, and 250 mg/kg/day-B.I.D. for 14 weeks (n = 15/group except where noted).

Organ/Tissue	0		5		25		125		250	
	F	M	F	M	F	M	F	M	F	M
Liver										
-hepatocyte hypertrophy	0	0	12	5	15	12	15	15	15	15
-diffuse vacuolation	5	4	6	3	7	4	10	6	6	11
Thyroid gland -follicular cell, diffuse hyperplasia	0	0	1	4	4	7	7	9	12	12
Pituitary gland										
-vacuolation	0	0	0	0	0	0	0	2	0	2
-cyst	0	0	0	0	0	0	0	0	4	0
Adrenal gland	n=15	n=15	n=1	n=2	n=1	n=0	n=0	n=1	n=15	n=15
-cortex,diffuse vacuolation	0	0							0	2

10. Plasma Drug Levels: Plasma C_{max} and AUC values for L-754,030 in male and female rats plateaued at doses ≥125 mg/kg/day. Plasma C_{max} and AUC values for L-754,030 in female rats were 3 to 4 times greater than corresponding values in male rats.
Toxicokinetic parameters for plasma levels of L-754,030 at week 13 in rats that received L-754,030 by oral gavage at dose of 5, 25, 125, and 250 mg/kg/day-B.I.D.

Dose,	C_{max}, µg/mL		T_{max}, hr		$AUC_{0\text{-}24hr}$, µg*hr/mL	
mg/kg/day-B.I.D.	Male	Female	Male	Female	Male	Female
5	0.334	0.560	8.00	8.00	2.31	6.81
25	0.325	0.905	8.00	8.00	5.04	13.7
125	0.385	1.62	8.00	16.00	6.04	27.3
250	0.333	1.64	24.00	8.00	6.37	25.7

Rats received L-754,030 by oral gavage at doses of 0, 5, 25, 125, and 250 mg/kg/ day-B.I.D. for 14 weeks. A no effect dose was not established. Target organs of toxicity were the liver, thyroid gland, and pituitary gland. For the liver, very slight to slight hepatocellular hypertrophy was observed in all male and female treatment groups. Hepatocyte hypertrophy most likely represents an induction of cytochrome P-450 enzymes and has no toxicological significance. Very slight to slight diffuse hepatocellular vacuolation was observed with an increased incidence for male rats that received a dose of 250 mg/kg/day-B.I.D. For the thyroid gland, very slight to slight diffuse thyroid follicular cell hyperplasia was observed for all male and female treatment groups. Follicular cell hyperplasia of the thyroid gland is most likely due to increased catabolism of thyroxine (T_4) and triiodothyronine (T_3) by the liver. For the pituitary gland, a very slight to slight vacuolation of individual cells in the pars distalis was observed for male rats that received doses of 125 and 250 mg/kg/day-B.I.D. This change was characterized by enlargement of individual pituitary cells due to formation of large cytoplasmic vacuoles and occasional protein droplets. This change may represent a degeneration or exhaustion of Thyroid stimulating hormone-producing pituitary cells secondary to hepatic enzyme induction and increased catabolism of T_3 and T_4. For the adrenal gland, diffuse vacuolation was observed in the cortex for male rats that received 250 mg/kg/day-B.I.D. Plasma C_{max} and AUC values for L-754,030 during week 13 in male and female rats plateaued at doses ≥125 mg/kg/day. Plasma C_{max} and AUC values for L-754,030 in female rats were 3 to 4 times greater than corresponding values in male rats.

Study title: Twenty Seven (27)-Week Oral Toxicity Study in Rats.

Key study findings: In the 27-week oral toxicity study with MK-0869 (— particle size) in Sprague-Dawley rats, treatment-related alterations of the hematology and clinical chemistry parameters were observed at all doses in both males and females. Hepatocellular hypertrophy and thyroid follicular cell hyperplasia were observed in both sexes in all treatment groups. The no effect dose was not identified, and the target organs of toxicity were the liver and the thyroid gland.

Study no: 01-092-0
Volume #, and page #: Vol #15, page # A4938

Conducting laboratory and location: Merck Research Laboratories, Merck & Co., Inc., West Point, PA.
Date of study initiation: July 11, 2001
GLP compliance: Yes
QA report: yes (X) no ( )
Drug, lot #, radiolabel, and % purity: Four batches of MK-0869 blended coated —— beads were used in the study. Batches #X0869OPP015C001 (also known as L-754030-016S001) and #F0869OPP018C004 (also known as L-754030-016S002) were obtained by blending 3 and 2 batches, respectively, of MK-0869 coated beads. Batches #X0869OPP023C001 (also known as L-754030-018W005) and #X0869OPP024C001 (also known as L-754030-018W006) were used without blending multiple batches of coated beads. The purity of the batches ranged from 99.6% to 100%.

Formulation/vehicle: Dosing formulations were prepared by dispersing the coated beads in 4% hydroxypropylcellulose, 20% sucrose and 0.19% sodium lauryl sulfate (SLS) in deionized water. The average particle size of MK-0869 in the dispersion was —— nm. Control 1 animals received 0.5% methylcellulose in deionized water and control 2 animals received 4% hudroxypropylcellulose, 20% sucrose and 0.19% SLS in deionized water.

Methods: There were two control groups in the study, and the treatment groups received 125, 500 and 1000 mg/kg b.i.d. doses of the drug. The second dose was administered approximately 6 hours after the first dose.

Dosing:

Species/strain: Crl:CD (SD)IGS BR Sprague-Dawley rats.
#/sex/group or time point (main study): 20 animals/sex/group were used in the study, except in the 500 mg/kg group which had 21 males and 19 females due to a gender identification error.
Satellite groups used for toxicokinetics or recovery: None
Age: 37 days old at study initiation.
Weight: Males: 122 to 178 g; Females: 87 to 147 g.
Doses in administered units: The treatment groups received the drug at 125, 500 and 1000 mg/kg b.i.d. (250, 1000 and 2000 mg/kg/day) doses.
Route, form, volume, and infusion rate: Dosing formulations were prepared by dispersing the drug-coated beads in 4% hydroxypropylcellulose, 20% sucrose and 0.19% sodium lauryl sulfate, and the doses were administered by oral gavage (5 ml/kg b.i.d).

Observations and times:

Clinical signs: The animals were observed daily for clinical signs and mortality.

Body weights: Body weights were recorded once prior to initiation of dosing and once or twice a week during the dosing period.

Food consumption: Food consumption was measured twice per week during weeks 1 to 13 and twice every 4th week thereafter.

Ophthalmoscopy: Ophthalmoscopic examinations were performed of all control 1, control 2 and high dose animals in weeks 12 and 26.

Hematology: Blood samples for hematological examinations were collected in dosing weeks 4, 12 and 25. The following hematological parameters were determined: erythrocytes, reticulocytes,

hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, platelets, leukocytes, leukocytes differential count and cell morphology.

Clinical chemistry: Blood samples for clinical chemistry analyses were collected in Drug Weeks 4, 12 and 25.

Urinalysis: Urinalysis was performed on urine samples collected from 10 rats/sex/group in Dosing Weeks 12 and 25.

Gross pathology: Complete necropsies of all animals were conducted at termination. Animals that died or were sacrificed moribund were also examined macroscopically.

Organs weighed: The weights of the following organs from all animals were recorded: adrenals, brain, heart, ovaries, kidneys, liver, pituitary, prostate, spleen, testes, thyroids.

Histopathology: The following tissues from the vehicle control, high dose and animals found dead/sacrificed early were examined histologically.
Salivary gland, esophagus, stomach, small intestine (duodenum, jejunum, ileum), large intestine, liver, pancreas, adrenals, parathyroid, pituitary, thyroid, kidneys, urinary bladder, ovaries, uterus, skin, mammary gland, lung, heart, spleen, lymph nodes (cervical and mesenteric), thymus, bone marrow, bone, skeletal muscle, brain, spinal cord, peripheral nerve, eye, testes and epididymides, prostate, harderian gland.

Toxicokinetics: Blood samples for toxicokinetic analysis were collected from 4 animals/sex/treatment group/sampling time at 2, 4, 6, 8, 10, 16 and 24 hours post-dose in Week 13. Plasma concentrations of MK-0869 were determined by [redacted]

Results:

Mortality: One high dose female was found dead in Week 4. The cause of the death was not known, however, it could be related to treatment with the drug. Another high dose female died from possible anesthesia accident in Week 25. One high dose male died in Week 12 and the cause of the death was stated to be an intubation accident. One mid-dose male was found dead following anesthesia in Week 4 and one low dose male was sacrificed because of trauma.

Clinical signs: No treatment-related clinical signs were observed in any group.

Body weights: The mean body weights of the control 1 and control 2 males before initiation dosing were 155±11 and 157±9 g and at the end of the dosing period were 485±20 and 522±26 g, respectively. The mean body weights of the control 1 and control 2 females before initiation of dosing were 124±8 and 117±10 g and at the end of dosing were 269±13 and 283±20 g, respectively. Treatment with MK-0869 was not associated with any changes in the body weights in any group.

Food consumption: The sponsor stated that there were no treatment-related changes in the food consumption any group (no data provided).

Ophthalmoscopy: No treatment-related abnormal ophthalmic changes were observed in any group.

Hematology: Increases in the platelet levels were observed in treatment group males at Weeks 4, 12 and 25 and in females at Week 4. In treatment group females, slight decreases in hemoglobin, hematocrit, MCV, MCH and MCHC were observed in Weeks 12 and 25. Hematological changes observed in the male and female animals at different times of the treatment period are shown in the Table below.

Parameter	Treatment Week	Control values	Percent changes from control		
			250 mg/kg/day	1000 mg/kg/day	2000 mg/kg/day
Males:					
Platelets (1,000/MM^3)	Week-4	1278 ± 161	+20	+15	+14
	Week-12	1247 ± 220	+18	+12	+14
	Week-25	1240 ± 163	+19	+17	+18
Females:					
Platelets (1,000/MM^3)	Week-4	1383 ± 167	+27	+23	+30
Hemoglobin (g/100 ml)	Week-12	14.4 ± 0.5	-8	-8	-8
	Week-25	14.0 ± 0.6	-11	-12	-11
Hematocrit (%)	Week-12	40.7 ± 1.3	-6	-6	-7
	Week-25	40.2 ± 1.7	-10	-10	-9
MCV (Cubic micron)	Week-12	53.0 ± 1.1	-4	-5	-5
	Week-25	53.8 ± 1.2	-6	-6	-6
MCH (micro gram)	Week-12	18.7 ± 0.4	-5	-7	-6
	Week-25	18.8 ± 0.5	-8	-8	-8
MCHC (g/dl)	Week-25	18.8± 0.5	-2	-3	-2

Clinical chemistry: There were increases in the serum protein and albumin levels, and decreases in albumin/globulin ratio in both males and females receiving MK-0869. Treatment group females had slight decreases in the alkaline phosphatase levels. Both males and females had slightly higher serum potassium and calcium levels. Treatment group females had higher cholesterol levels and both males and females had lower triglyceride levels. Males also had slightly higher cholesterol levels during Week 12. However, none of the effects were dose-dependent, except the cholesterol levels in the males. The changes in the clinical chemistry parameters in the male and female animals are summarized in the Table below.

Parameter	Treatment Week	Control values	Percent changes from control		
			250 mg/kg/day	1000 mg/kg/day	2000 mg/kg/day
Males:					
Protein (g/dL)	Week-4	5.6 ± 0.2	+4	+7	+5
	Week-12	6.2 ± 0.2	+6	+10	+6
	Week-25	6.5 ± 0.2	+8	+9	+8
Albumin (g/dL)	Week-12	3.5 ± 0.1	+3	+6	+3
	Week-25	3.5 ± 0.2	+6	+6	+6
A/G Ratio	Week-4	1.4 ± 0.1	-14	-14	-14
Potassium (meq/L)	Week-4	5.1 ± 0.3	+6	+8	+8
	Week-12	4.9 ± 0.2	+8	+6	+6
	Week-25	5.0 ± 0.2	+10	+12	+10
Cholesterol (mg/dL)	Week-12	64 ± 9	+20	+28	+29
Triglycerides (mg/dL)	Week-4	65 ± 36	-48	-48	-55
	Week-12	96 ± 43	-56	-61	-63
	Week-25	88 ± 41	-65	-69	-75
Females:					
Protein (g/dL)	Week-4	6.0 ± 0.3	+12	+16	+12
	Week-12	6.6 ± 0.3	+19	+23	+19
	Week-25	7.1 ± 0.6	+19	+20	+17
Albumin (g/dL)	Week-12	3.8 ± 0.2	+11	+16	+11
	Week-25	4.2 ± 0.4	+9	+12	+7
A/G Ratio	Week -4	1.6 ± 0.1	-19	-19	-25
	Week-12	1.4 ± 0.1	-14	-7	-14

	Week-25	1.5 ± 0.2	-19	-19	-19
Alkaline Phosphatase (U/L)	Week-4	146 ± 30	-20	-20	-20
	Week-12	67 ± 14	-21	-23	-15
	Week-25	45 ± 18	-23	-17	-9
Potassium (meq/L)	Week-4	4.6 ± 0.2	+9	+4	+7
	Week-12	4.5 ± 0.2	+9	+9	+9
	Week-25	4.4 ± 0.3	+7	+12	+12
Cholesterol (mg/dL)	Week-4	83 ± 10	+50	+68	+59
	Week12	79 ± 11	+85	+105	+92
	Week25	97 ± 20	+88	+107	+88
Triglycerides (mg/dL)	Week-4	66 ± 24	-13	-26	-43
	Week-12	56 ± 19	-17	-17	-37
	Week-25	80 ± 37	-30	-16	-37

Urinalysis: No significant treatment-related changes were observed in any group.

Organ weights: Treatment-related increases in the liver and thyroid weights (both absolute and relative) were observed in both males and females receiving MK-0869. The increases in the liver and thyroid weights were not dose-related and appeared to plateau between 250 and 1000 mg/kg/day doses. The liver and thyroid weights (in grams) and the changes in absolute and relative weights in different groups of animals are summarized in the Table below.

Organs	Control Weights		Percent Changes From Control					
			250 mg/kg/day		1000 mg/kg/day		2000 mg/kg/day	
	Males	Females	Males	Females	Males	Females	Males	Females
Liver (absolute)	12.73±0.97 g	8.04±0.74 g	57%	97%	70%	116%	65%	107%
Liver (relative)	2.70±0.18	3.09±0.27	58%	98%	72%	120%	67%	112%
Thyroid (absolute)	0.024±0.0033 g	0.014±0.0016 g	60%	56%	61%	74%	44%	74%
Thyroid (relative)	0.0051±0.0007	0.0056±0.0006	61%	56%	63%	77%	46%	77%

Gross pathology: Males and females of all treatment groups had enlarged liver and thyroid glands. The enlargement of the liver was associated with hepatocellular hypertrophy, characterized by enlarged eosinophilic hepatocytes surrounding the central vein.

Histopathology: Hepatocellular hypertrophy, characterized by enlarged eosinophilic hepatocytes was observed in all treatment group animals. In the mid- and the high-dose groups, enlarged hepatocytes extended into the midzonal region. The severity of the hepatocellular hypertrophy appeared to plateau at 500 mg/kg b.i.d. in females and 125 mg/kg b.i.d. in males. Individual hepatocytes, in all dose groups, underwent single cell necrosis and had increased mitotic activity. Diffuse thyroid follicular cell hypertrophy, characterized by tall columnar follicular cells with an expanded cytoplasm was observed in both males and females of all treatment groups. The incidences of histopathological changes observed in the liver and thyroid glands of males and females of different groups are shown in the sponsor's Table below.

Histomorphologic Changes
(Incidence, n=20)

	MK-0869 (mg/kg b.i.d.)									
	Females					Males				
	Control 1	Control 2	125	500	1000	Control 1	Control 2	125	500	1000
Liver										
Eosinophilic cellular alteration	1	-	1a	1a	-	-	-	2a	-	1a
Centrilobular hypertrophy	-	-	20a	20a	20a	-	-	20a	20a	20a
Midzonal vacuolation	-	-	8a	12a	9a	-	-	20a	19a	20a
Single-cell necrosis	3	-	12a	16a	17a	3	3	19a	19a	18a
Mitotic activity	3	-	10a	13a	8a	-	1	5a	9a	5a
Multinucleated hepatocytes	-	-	17a	16a	17a	-	-	2a	4a	2a
Thyroid										
Hypertrophy, diffuse, follicular cell	-	-	19a	19a	19a	-	-	20a	19a	20a
Hyperplasia, Focal, cystic, follicular cell	-	-	-	-	-	-	-	2a	-	-

[a] Treatment-related changes based on incidence and/or severity
- Not observed.

Toxicokinetics: Following multiple dosing to male and female rats, the maximum plasma concentration (C_{max}) and the plasma exposure (AUC) values were up to 4-fold higher in females as compared to that of males. The C_{max} was achieved in 8 to 12 hours after dosing in both sexes. The exposure levels appeared to plateau at the 250 mg/kg bi.d. dose in both males and females. Plasma toxicokinetic parameters in male and female animals receiving MK-0869 are shown in the sponsor's Table below.

Mean Plasma MK-0869 Toxicokinetic Parameters - Drug Week 13

	MK-0869 (mg/kg b.i.d.)		
	Females		
	125	500	1000
$AUC_{0-24\ hr}$ (μg•hr/mL)[a]	27.9 ± 3.12	26.0 ± 2.74	31.3 ± 2.22
C_{max} (μg/mL)[b]	1.84	1.57	1.97
T_{max} (hr)[c]	10	8.0	10
	Males		
	125	500	1000
$AUC_{0-24\ hr}$ (μg•hr/mL)[a]	6.51 ± 0.386	7.38 ± 0.546	8.02 ± 0.588
C_{max} (μg/mL)[b]	0.456	0.410	0.511
T_{max} (hr)[c]	10	10	10

[a] Mean ± SEM calculated using all individual plasma concentrations.
[b] Maximum mean plasma concentrations.
[c] Time at which C_{max} occurred

Summary: In the 27-week oral toxicity study MK-0869 in Sprague-Dawley rats, groups of animals received 125, 500 and 1000 mg/kg b.i.d. (250, 1000 and 2000 mg/kg/day) doses of the drug.

Treatment-related changes in the hematology (increased platelet levels in males and increased platelets, decreased hemoglobin and hematocrit values in females) and clinical chemistry (increased protein and cholesterol and decreased triglycerides in both sexes) parameters were observed in all treatment group animals. Hepatocellular hypertrophy and thyroid follicular cell hyperplasia were observed in both males and females of all treatment groups. Thus, the target organs of toxicity were the liver and the thyroid glands, and the no effect dose was not established.

53-Week Oral Toxicity Study in Rats with a 27-Week Interim Necropsy (TT #97-071-0).

Testing Laboratory: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486

Date Started: July 7, 1997

Date Completed: November 18, 1998

GLP Compliance: Statements of compliance with GLP regulations and the Quality Assurance Unit were included.

Animals: Sprague-Dawley rats [Crl:CD®(SD)IGS BR] were used in this study. At the start of treatment, animals were 35 days of age and had body weight ranges of 100 to 174 g for male rats and 93 to 130 g for female rats.

Drug Batch: L-754,030-004H021 and L-754,030-004H026 — drug particle size)

Methods: In a 52-week oral toxicity study, rats received L-754,030 (— drug particle size) at doses of 0, 0.25, 25, and 250 mg/kg/day. There were 30 rats/sex/group. An interim evaluation was conducted with 10 rats/sex/group, which were sacrificed after treatment for 26 weeks. For animals sacrificed at 27 weeks for interim evaluation, male and female rats received 182 and 183 doses, respectively. For animals sacrificed at the termination of the study at 53 weeks, male and female rats received 364-365 and 365-366 doses, respectively. Control animals received the vehicle, 0.5% aqueous methylcellulose containing 0.02% sodium lauryl sulfate. Vehicle or drug suspension was administered by oral gavage using a dose volume of 5 mL/kg. Animals were observed daily for clinical signs of toxicity and moribundity/mortality. Body weight was measured once in week 1, twice per week through week 13, and once per week thereafter. Food consumption was evaluated twice per week from weeks 1 through 13 and twice per week every 4 weeks thereafter. Ophthalmic examinations were conducted in all surviving animals from the control and 250 mg/kg/day groups during weeks 12, 24, 39, and 52. Blood for determination of hematology and serum biochemical parameters was collected during weeks 4, 12, and 25 from 9 to 10 rats/sex/group (rats designated for the interim necropsy) and during weeks 39 and 51 from all surviving animals (rats designated for the final necropsy). Urinalysis was conducted during weeks 12 and 25 with 7 to 10 rats/sex/group (rats designated for interim sacrifice) and during weeks 39 and 51 with surviving animals (rats designated for the final necropsy). At the scheduled interim sacrifice (i.e., week 27), 9 or 10 rats/sex/group were sacrificed, and at the scheduled terminal sacrifice

(i.e., week 53), 19 or 20 rats/sex/group were sacrificed. Complete gross examinations were conducted on all animals, including those that died or were sacrificed in a moribund condition during the treatment period prior to scheduled necropsy. Absolute and relative organ weights were determined for the adrenal gland, brain, heart, ovaries, kidneys, liver, pituitary gland, prostate, spleen, testes, and thyroid gland. Organs and tissues were collected and preserved from all animals as follows: lung, heart, liver, kidneys, urinary bladder, spleen, thymus, lymph nodes, adrenal glands, thyroid gland, parathyroid gland, pituitary gland, salivary gland, stomach, small intestine, esophagus, pancreas, spinal cord, peripheral nerve, eye (with optic nerve and Harder's gland), skin (from mammary region), bone, bone marrow, testes and epididymides, prostate, ovaries, uterus, brain, skeletal muscle, and large intestine. Complete histopathological examinations were conducted on all animals in the control and 250 mg/kg/day groups sacrificed at interim and final necropsies and from all animals which died or were sacrificed in a moribund condition prior to scheduled termination. Further all gross changes and sections of liver and thyroid gland were submitted to histopathological examination from all animals.

Results:

1. Observed Effects: No treatment-related observed effects.

2. Mortality: Death or moribund sacrifice during the treatment period occurred for 1 female rat at 0.25 mg/kg/day, 1 female rat at 25 mg/kg/day, and 1 male rat and 3 female rats at 250 mg/kg/day. None of the deaths or moribund sacrifices were considered to be treatment-related.

Death and moribund sacrifice during the treatment period.

Animal #	Dose, mg/kg/day	Number of doses	Death/ Moribund Sacrifice	Cause of Death/ Moribund Sacrifice
97-4201F	25	29	Moribund Sacrifice	Urinary obstruction attributed to pyelonephritis
97-4294M	250	67	Moribund Sacrifice	Bladder filled with blood and markedly enlarged kidney (i.e., pyelonephritis)
97-4275F	250	97	Moribund Sacrifice	Intubation accident
97-4247F	250	309	Moribund Sacrifice	Gastrointestinal distention
97-4123F	0.25	320	Death	Esophageal impaction with food
97-4265F	250	351	Moribund Sacrifice	Pituitary adenoma

3. Body Weight and Food Consumption: There were no treatment-related effects on body weight gain, final body weight, or food consumption. Mean body weight of male controls prior to the start of treatment and at weeks 26 and 52 were 137, 471, and 531 g, respectively. Mean body weights of female controls prior to the start of treatment and at weeks 26 and 52 were 111, 257, and 282 g, respectively.

Body weight gain and final body weight, expressed as a percentage of the control, for male and female rats that received L-754,030 at oral doses of 0, 0.25, 25, and 250 mg/kg/day for 26 or 52 weeks.

Week	0.25 mg/kg/day		25 mg/kg/day		250 mg/kg/day	
	Male	Female	Male	Female	Male	Female
Week 26						
-body weight gain (wk 0-26), % of control	98.4	96.4	98.6	103.7	100.3	98.9
-body weight, % of control	99.6	98.8	100.4	98.8	100.2	101.2
Week 52						
-body weight gain (wk 0-52), % of control	98.8	95.6	98.8	97.9	100.25	95.3
-body weight, % of control	100.4	97.9	100.6	98.2	100	98.9

4. **Hematology:** For female rats at 250 mg/kg/day, there were slight decreases of hemoglobin levels (93-97.2% of the control, 13.4-14.3 g/100 mL) and hematocrit (92.8-96.8% of the control, 38.4-40.6%) at weeks 4, 12, 25, 39, and 51; however, the biological significance of these changes is questionable.

5. **Blood Biochemistry and Urinalysis:** Total cholesterol levels were increased for female rats at 25 and 250 mg/kg/day during weeks 4, 12, 25, 39, and 51. Triglyceride levels were decreased for male rats at 250 mg/kg/day during weeks 4, 12, 25, 39, and 51, male rats at 25 mg/kg/day during weeks 12, 25, 39, and 51, and male rats at 0.25 mg/kg/day during weeks 39 and 51. There were no treatment-related changes of urinalysis parameters.

Male rats: Triglyceride levels for male rats at 250 mg/kg/day during weeks 4, 12, 25, 39, and 51 were decreased to 72.7, 79.1, 80.3, 55.9, and 50.4% of the control (44-131 mg/dL), respectively. Triglyceride levels for male rats at 25 mg/kg/day during weeks 12, 25, 39, and 51 were decreased to 91.0, 90.9, 61.3, and 55% of the control, respectively. Triglyceride levels for male rats at 0.25 mg/kg/day during weeks 39 and 51 were decreased to 82 and 76.3% of the control, respectively.

Female rats: Total cholesterol levels for female rats at 25 mg/kg/day during weeks 4, 12, 25, 39, and 51 were increased to 111.7, 117.65, 124.4, 130.3, and 126% of the control (68-96 mg/dL), respectively. Total cholesterol levels for female rats at 250 mg/kg/day during weeks 4, 12, 25, 39, and 51 were increased to 116.9, 127.9, 126.9, 136, and 127% of the control, respectively. Triglyceride levels for female rats at 25 and 250 mg/kg/day during week 4 were decreased to 81.5 and 74.1% of the control (54 mg/dL), respectively; however, no changes were observed at later time points in the study. Alanine aminotransferase activities for female rats at 25 and 250 mg/kg/day during week 51 were increased to 129.3 and 148.3% of the control (58 U/L), respectively. Aspartate aminotransferase activity for female rats at 250 mg/kg/day was increased to 116.7% of the control (108 U/L). Albumin/globulin ratios for female rats at 250 mg/kg/day during weeks 4, 12, 25, 39, and 51 were decreased to 86.7, 92.3, 84.6, 91.7, and 92.3% of the control (1.2-1.5), respectively.

6. **Ophthalmic Examinations:** Ophthalmic examinations conducted in all surviving animals from the control and 250 mg/kg/day groups during weeks 12, 24, 39, and 52 revealed no apparent treatment-related findings. The sponsor provided no data for independent analysis.

7. **Organ Weights:** Changes in liver and thyroid gland weights were observed that appeared to correlate with histopathological findings.

Liver: At interim necropsy (week 27), absolute liver weights for male rats at 25 and 250 mg/kg/day were increased to 118.5 and 123.5% of the control (11.79 g), respectively. Relative liver weight for male rats at 25 and 250 mg/kg/day were increased to 120 and 126.2% of the control (2.60% B.W.), respectively. Absolute liver weights for female rats at 25 and 250 mg/kg/day were increased to 131.35 and 145.25% of the control (7.05 g), respectively. Relative liver weights for female rats at 25 and 250 mg/kg/day were increased to 126.7 and 143.4% of the control (2.88%), respectively. At terminal necropsy (week 53), absolute liver weights for male rats at 25 and 250 mg/kg/day were increased to 116.9 and 128% of the control (12.95 g), respectively. Relative liver weights for male rats at 25 and 250 mg/kg/day were increased to 117.2 and 129% of the control (2.62% B.W.), respectively. Absolute liver weights for female rats at 25 and 250 mg/kg/day were increased to 126.9 and 142.9% of the control (7.46 g), respectively. Relative liver weights for female rats at 25 and 250 mg/kg/day were increased to 127.3 and 144.2% of the control (2.85% B.W.), respectively.

Thyroid gland: At interim necropsy (week 27), absolute and relative thyroid gland weights for male rats at 250 mg/kg/day were increased to 114.6 and 117% of control values (0.0275 g and 0.0062% B.W.), respectively. Absolute and relative thyroid gland weights for female rats at 250 mg/kg/day were increased to 120 and 118% of control values (0.0210 g and 0.0085% B.W.), respectively. At terminal necropsy (week 53), absolute and relative thyroid glands for male rats at 250 mg/kg/day were increased to 109.6 and 111.3% of control values (0.0286 g and 0.0059% B.W.), respectively. Absolute thyroid gland weights for female rats at 25 and 250 mg/kg/day were increased to 110.7 and 117.7% of the control (0.0187 g), respectively. Relative thyroid gland weights for female rats at 25 and 250 mg/kg/day were increased to 111.1 and 118.1% of the control (0.0072% B.W.), respectively.

8. Gross Pathology: Not reported.

9. Histopathology: Target organs of toxicity were the liver and thyroid gland following treatment for either 26 or 52 weeks. For the liver, slight centrilobular hypertrophy was observed for both male and female rats at 25 and 250 mg/kg/day following treatment for 26 or 52 weeks. For the thyroid gland, slight diffuse follicular cell hyperplasia was observed for both male and female rats at 25 and 250 mg/kg/day following treatment for 26 or 52 weeks. For the liver, organ weight and histological changes appear to be consistent with an induction of cytochrome P-450.

Histopathological findings for rats that received L-754,030 by the oral route at doses of 0, 0.25, 25, and 250 mg/kg/day for 26 weeks (Interim Sacrifice).

Organ/Tissue	0 mg/kg/day		0.25 mg/kg/day		25 mg/kg/day		250 mg/kg/day	
	Female	Male	Female	Male	Female	Male	Female	Male
Liver, n =	10	10	10	10	11	10	11	10
-hemorrhage	0	0	0	0	0	0	0	1
-centrilobular hypertrophy	0	0	0	0	6	6	10	10
-focal necrosis	0	0	0	0	0	0	1	2
Thyroid gland, n =	10	10	10	10	11	10	11	10
-follicular cell diffuse hyperplasia	0	0	0	0	5	5	10	8
Pituitary gland, n =	10	10	1	-	1	-	11	10
-hyperplasia	1	-	-	-	-	-	2	1
Kidney, n =	10	10	1	-	1	1	11	10
-transitional epithelium, hyperplasia	-	-	1	-	-	-	-	2
-mineralization	1	-	1	-	-	-	3	3
-pyelonephritis	-	-	-	-	1	-	-	1

Histopathological findings for rats that received L-754,030 by the oral route at doses of 0, 0.25, 25, and 250 mg/kg/day for 53 weeks (Terminal Sacrifice).

Organ/Tissue	0 mg/kg/day		0.25 mg/kg/day		25 mg/kg/day		250 mg/kg/day	
	Female	Male	Female	Male	Female	Male	Female	Male
Liver, n =	20	20	20	20	19	20	19	20
-centrilobular hypertrophy	0	0	0	0	16	13	18	19
-focal necrosis	-	-	-	-	-	-	1	2
Thyroid gland, n =	20	20	18	20	19	20	19	20
-follicular cell diffuse hyperplasia	-	-	-	-	2	2	10	12
-follicular cell, focal cystic hyperplasia	-	-	-	-	1	-	-	-
Adrenal gland, n=	20	20	4	1	4	1	19	20
-benign pheochromocytoma	-	-	-	-	-	-	-	1
Parathyroid gland, n=	20	20	1	-	-	-	19	20
-benign adenoma	-	-	-	-	-	-	0	1
Pituitary gland, n =	20	20	6	1	3	2	19	20
-benign adenoma	1	1	-	-	1	2	1	1
Large Intestine, n =	20	20	1	1	-	-	19	20
-rectum, malignant leiomyosarcoma	-	-	-	1	-	-	-	-
Skin, n =	20	20	1	-	-	-	19	20
-benign papilloma	-	-	-	-	-	-	-	1

In a 52-week oral toxicity study, rats received L-754,030 (— drug particle size) at doses of 0, 0.25, 25, and 250 mg/kg/day. An interim evaluation was conducted after treatment for 26 weeks. The no effect dose was 0.25 mg/kg/day. There was no treatment-related mortality. Target organs of toxicity were the liver and thyroid gland following treatment for either 26 or 52 weeks. For the liver, slight centrilobular hypertrophy was observed for both male and female rats at 25 and 250 mg/kg day following treatment for 26 or 52 weeks. For the thyroid gland, slight diffuse follicular cell hyperplasia was observed for both male and female rats at 25 and 250 mg/kg/day following treatment for 26 or 52 weeks. For the liver, increased absolute and relative organ weights as well as histological changes appeared to be consistent with an induction of cytochrome P-450. This study used the — drug particle size and selection of the high dose was based upon a saturation of absorption for the parent compound. Subsequent toxicokinetic studies suggested that observed saturation of absorption was actually a saturation of dissolution related to the drug particle size as exposure (i.e., AUC) was found to increase with a — drug particle size formulation. The sponsor is conducting a 5 week oral toxicity/toxicokinetic study in rats designed as a bridging study to link the — drug particle size formulations as communicated in Amendment #115 submitted on January 7, 2000. If this bridging study fails to demonstrate saturation for the — particle size formulation, dose selection for this 52-week oral toxicity study may be deemed to be inadequate and the study might have to be repeated.

Mice

Five-Week Oral Range Finding Study in Mice (Report Date/Number TT #97-069-0).

Testing Laboratory: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486

Study Started: July 17, 1997

Study Completed: January 27, 1998

GLP Compliance: Statements of Compliance with GLP Regulations and the Quality Assurance Unit were included.

Animals: Crl:CD-1®(ICR)BR mice were used in this study. At the start of treatment, animals were 44 days old and body weight ranges were 24.3-33.7 g for male mice and 20.6-28.9 g for female mice.

Drug Batch: L-754,030-004H21

Methods: Mice received L-754,030 by the oral route of administration at doses of 0 and 500 mg/kg/day or at doses of 0, 500, and 1000 mg/kg/day B.I.D. (total daily doses of 0, 1000, and 2000 mg/kg, respectively) for 28 (female mice) or 29 (male mice) days. Mice in the B.I.D. control, 500 mg/kg/day, and 1000 mg/kg/day groups were dosed twice per day, with second dose administered 6 hr after the initial dose. Mice in control groups received the vehicle, 0.5% methylcellulose/0.02% sodium lauryl sulfate. There were 12 mice/sex/group. The dosing volume was 10 mL/kg. Mice were observed daily for mortality and clinical signs of toxicity. Body weights were measured once prior to the start of treatment, once in week 1, and twice per week in weeks 2 to 4. Group food consumption (n = 3 mice/box) was measured over 4 days each week. Ophthalmic examinations were performed on all animals prior to the start of treatment and on all animals in control B.I.D. and 1000 mg/kg/day B.I.D. groups at week 4. Blood for determination of hematological and biochemical parameters was collected from 12 mice/sex/group immediately prior to necropsy in week 5. Necropsy examinations were performed on all animals. Absolute and relative organ weights were determined for the brain, spleen, heart, kidneys, liver, and testes. Tissues from 5 mice/sex in the control B.I.D. and 1000 mg/kg/day B.I.D. groups were collected, processed, and microscopically examined as follows: salivary gland, esophagus, stomach, small intestine, large intestine, liver, gall bladder, pancreas, adrenal glands, pituitary glands, thyroid gland, parathyroid gland (when present in the thyroidal section), kidneys, urinary bladder, ovaries, testes (to include epididymis), uterus, prostate, heart, lung, thymus, lymph node, spleen, skin, from mammary region, mammary gland (when present in skin section), bone (with marrow), skeletal muscle, brain (to include cerebral cortex, and subcortical white matter, thalamus, hypothalamus, mid-brain, cerebelum, pons, and medulla), cervical spinal cord, peripheral

nerve (sciatic), eye (to include optic nerve and Harderian gland). In addition, any gross abnormalities were examined.

Results:

1. Observed Effects: The sponsor did not report any treatment-related observed effects.

2. Mortality: **There was no treatment-related mortality.**

3. Body Weight and Food Consumption: Body weight gains were impaired by >10% for the female 500 and 1000 mg/kg/day B.I.D. groups. Body weight gain for the male 500 mg/ kg/day group was also impaired by >10%; although, this change may be unrelated to treatment due to lack of change in the male 500 and 1000 mg/kg/day B.I.D. groups. Body weights for the male control group at weeks –1 and 4 were 24.7 and 29.0 g, respectively. Body weight gain for the male 500 mg/kg/day group was reduced to 77.9% of the control. Food consumption for the male 500 mg/kg/day group was reduced to 94.3% of the control (5.3 g/animal/day). Body weights for male control B.I.D. group at weeks –1 and 4 were 23.8 and 26.1 g, respectively. Body weight gains for male 500 and 1000 mg/kg/day B.I.D. groups were 112.7 and 96.7% of the control, respectively. Body weights for the female control group at weeks –1 and 4 were 29.1 and 32.7 g, respectively. Body weight gain for the female 500 mg/kg/day group was 100.4% of the control. Body weights for female control B.I.D. group at weeks –1 and 4 were 28.6 and 31.9 g, respectively. Body weight gains for female 500 and 1000 mg/kg/day B.I.D. groups were 60.7 and 75.9% of the control, respectively.

4. Hematology: Leukocyte and lymphocyte counts for the male 1000 mg/kg/day B.I.D. groups were both increased to 129% of the control (2450 cells/mm^3 and 1953 cells/mm^3).

5. Blood Biochemistry and Urinalysis: Cholesterol levels for the male 500 and 1000 mg/ kg/day B.I.D. groups were increased to 126.5 and 121.7% of the control (83 mg/dL), respectively. Cholesterol levels for the female 500 and 1000 mg/kg/day B.I.D. groups were increased to 105.4 and 117.8% of the control (129 mg/dL), respectively. Triglyceride levels for the male 500 and 1000 mg/kg/day B.I.D. groups were increased to 132.2 and 154.8% of the control (62 mg/dL), respectively. Triglyceride levels for the female 500 and 1000 mg/ kg/day B.I.D. groups were increased to 152.7 and 174.5% of the control (55 mg/dL), respectively.

6. Ophthalmic Examination: There were no treatment-related ophthalmic effects; although, data was not presented for independent analysis.

7. Organ Weights: Liver weights were increased for male 500 and 1000 mg/kg/day B.I.D. groups and all female treatment groups; however, there were no corresponding histopathological changes. Absolute liver weights for the male 500 and 1000 mg/kg/day B.I.D. groups were increased to 115.3 and 120.9% of the control (1.1326 g), respectively. Relative liver weights for the male 500 and 1000 mg/kg/day B.I.D. groups were increased to 112.5 and 119.7% of the control (4.57%), respectively. Absolute and relative liver weights for the female 500 mg/kg/day group were increased to 109.7 and 107.3% of the control (1.2980 g and 4.36%), respectively. Absolute liver weights for the female 500 and 1000 mg/kg/day B.I.D. groups were increased to 109.3 and 118.9% of the control

(1.2429 g), respectively. Relative liver weights for the male 500 and 1000 mg/kg/day B.I.D. groups were increased to 113.6 and 117.1% of the control (4.26%), respectively.

8. Gross Pathology: The sponsor did not report any treatment-related gross pathological effects.

9. Histopathology: There were no treatment-related histopathological effects.

Mice received L-754,030 by the oral route of administration at doses of 0 and 500 mg/kg/day or at doses of 0, 500, and 1000 mg/kg/day B.I.D. (total daily doses of 0, 1000, and 2000 mg/kg, respectively) for 28 (female mice) or 29 (male mice) days. Mice in the B.I.D. control, 500 mg/kg/day, and 1000 mg/kg/day groups were dosed twice per day, with second dose administered 6 hr after the initial dose. The dose of 500 mg/kg/day could be considered a no effect dose. Body weight gains were impaired by >10% for the female 500 and 1000 mg/kg/day B.I.D. groups. Liver weights were increased for male 500 and 1000 mg/kg/day B.I.D. groups and all female treatment groups; however, there were no corresponding histopathological changes. Elevations of cholesterol and triglyceride levels were evident for male and female mice that received L-754,030 at doses of 500 and 1000 mg/kg/day B.I.D. A target organ of toxicity was not identified.

Study Title: MK-0869: Five-Week Oral Toxicity and Toxicokinetic Study in Mice.

Study No: TT # 99-095-0,-1 and TT # 99-087-0,-1
Conducting laboratory and location: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486
Date Started: November 8, 1999 (Toxicokinetic Study) and December 9, 1999 (Toxicology Study)
Date Completed: August 1, 2000 (Toxicokinetic Study) and August 3, 2000 (Toxicology Study)
GLP compliance: Statements of compliance with GLP regulations were included for the both toxicology and toxicokinetic studies.
QA- Report Yes (X) No ( )
Methods: The toxicology and toxicokinetic profiles of two MK-0869 formulations of different particle size, were assessed in CD-1 mice during a 5-week treatment period (28 to 29 days). MK-0869 Formulation M (average particle size, —) was administered to mice at an oral dose of 500 mg/kg/day. In the toxicology study, a control group received the vehicle, 5% methylcellulose and 0.02% sodium lauryl sulfate in deionized water (i.e., vehicle for Formulation M). MK-0869 Formulation NB (average particle size, —) was administered using S.I.D. and B.I.D. regimens. For the S.I.D. regimen, MK-0869 Formulation NB was administered at doses of 25, 500, 1000, 1250, and 1500 mg/kg/day. For the B.I.D. regimen, MK-0869 Formulation NB was administered at doses of 12.5, 250, 500, 625, and 750 mg/kg B.I.D. (total daily doses of 25, 500, 1000, 1250, and 1500 mg/kg, respectively). Animals dosed twice daily received the second dose approximately 6 hr following the first dose. In the

toxicology study, a control group received the vehicle, 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water twice per day (i.e., vehicle for Formulation NB).

Design of toxicology and toxicokinetic studies.

Formulation M			Formulation NB			Formulation NB		
Dose	mice/sex/group		Dose	mice/sex/group		Dose	mice/sex/group	
mg/kg/day	MS	TK	mg/kg/day	MS	TK	mg/kg B.I.D.	MS	TK
0 (MC/SLS)	10	-	0 (HPC/SLS/S)	10	-	0 (HPC/SLS/S)	10	-
500	15	30	25	15	30	12.5	15	30
			500	15	30	250	15	30
			1000	15	30	500	15	30
			1250	15	30	625	15	30
			1500	15	30	750	15	30

MS = main study and TK= toxicokinetic study.

Dosing:

- **species/strain:** Crl:CD-1®(ICR) BR mice were obtained from [redacted]

- **#/sex/group or time point:** Control groups were composed of 10 mice/sex/group and treatment groups were composed of 15 mice/sex/group.

- **age:** For the toxicology study, male and female mice were 49 and 44 days old, respectively, at the start of treatment. For the toxicokinetic study, male and female mice were 46 and 43 days old, respectively, at the start of treatment.

- **weight:** For the toxicology study, body weight ranges were 23.3 to 41.8 g for male mice and 19.2 to 29.5 g for female mice. For the toxicokinetic study, body weight ranges were 25.6 to 42.1 g for male mice and 18.2 to 29.0 g for female mice.

- **satellite groups used for toxicokinetics or recovery:** For the toxicokinetic study, there were 30 mice/sex/group.

- **dosage groups in administered units:** MK-0869 Formulation M was administered at doses of 0 and 500 mg/kg/day. MK-0869 Formulation NB was administered using S.I.D. and B.I.D. regimens. For the S.I.D. regimen, MK-0869 Formulation NB was administered at doses of 0, 25, 500, 1000, 1250, and 1500 mg/kg/day. For the B.I.D. regimen, MK-0869 Formulation NB was administered at doses of 0, 12.5, 250, 500, 625, and 750 mg/kg B.I.D. (total daily doses of 0, 25, 500, 1000, 1250, and 1500 mg/kg/day, respectively).

- **route, form, volume, and infusion rate:** Vehicle or drug suspension was administered by oral gavage using a dose volume of 10 mL/kg.

Drug, lot#, radiolabel, and % purity: MK-0869 Formulation M, lot number L-754030-004H031, had an average particle size of — with 99.5% purity. MK-0869, Formulation NB (MK-0869 blended coated — beads), batch #X0869OPP015C001 (also known as L-754030-016S001) with an average particle size of — was obtained by blending 3 batches of MK-0869 coated beads with purity ranging from 99.6 to 100.0%.

Formulation/vehicle: The vehicle for MK-0869 Formulation M was 5% methylcellulose and 0.02% sodium lauryl sulfate in deionized water. The vehicle for MK-0869 Formulation NB was 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water.

Observations and times:

- **Clinical signs:** Animals were monitored daily for clinical signs of toxicity and mortality before and after dosing.
- **Body weights:** Body weights were measured prior to the start of dosing, once during week 1, and twice per week, thereafter.
- **Food consumption:** Food consumption was measured once each week over a 4-day period.
- **Ophthalmoscopy:** Ophthalmic examinations, consisting of indirect ophthalmic examinations on all animals and slit lamp biomicroscopsy on animals at the discretion of the examiner, were conducted prior to the start of treatment. During week 4, ophthalmic examinations were conducted on all control animals and all animals in the 500 mg/kg/day MK-0869 Formulation M, 1500 mg/kg/day MK-0869 Formulation NB, and 750 mg/kg B.I.D. MK-0869 Formulation NB groups.
- **EKG:** Not performed.
- **Hematology:** Blood for determination of hematology parameters was collected from all animals at scheduled necropsy.
- **Clinical chemistry:** Blood for determination of clinical chemistry parameters was collected from all animals at scheduled necropsy.
- **Urinalysis:** Not performed.
- **Gross pathology:** Surviving mice at scheduled termination were sacrificed and submitted to necropsy examinations. Mice that died or were sacrificed in a moribund condition during the treatment period were also submitted to necropsy examinations.
- **Organs weighed:** Absolute and relative organ weights were determined for the brain, heart, kidneys, liver, spleen, and testes.
- **Histopathology:** Tissue sections from the control group that received 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water once per day, the control group that received 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water twice per day, the 1500 mg/kg/day Formulation NB group, and the 750 mg/kg B.I.D. Formulation NB group were prepared, stained with hematoxylin and eosin, and submitted to microscopic examination. Tissue sections from animals that died or were sacrificed in a moribund condition during the treatment period were also submitted to microscopic examination. Liver sections from all control and treatment groups were submitted to microscopic examination. Tissues examined were as follows: salivary gland (submandibular/sublingual), esophagus, stomach (glandular and nonglandular portions), small intestine (duodenum, jejunum, ileum), large intestine (colon), liver, gallbladder, pancreas, adrenal glands, parathyroid (when present in thyroidal sections), pituitary gland, thyroid gland, kidneys, urinary bladder, ovaries, prostate, skin (from mammary region), mammary gland (when present in skin section), lung, heart, spleen, lymph nodes (cervical and mesenteric), thymus, bone marrow (in bone section), bone (femur, including femorotibial joint), skeletal muscle, brain (including cerebral cortex and subcortical white matter, basal ganglia, cerebellum, and pons), spinal cord (cervical), peripheral nerve (sciatic), uterus, testes and epididymides, eye (with optic nerve), and Harder's gland. Tissues with gross lesions were also submitted to microscopic examination at the discretion of the pathologist.
- **Toxicokinetics:** Blood for determination of plasma drug levels was collected from 4 mice/sex/group/time point on day 29 or 30 at 2, 4, 6, 8, 10, 16, and 24 hour following the first daily dose. For the B.I.D. regimen, mice scheduled for the 8, 10, 16, and 24 hr time points,

received their second daily dose at approximately 6 hr after the first dose. Following scheduled blood collections, mice were sacrificed and discarded without examination. Two additional mice/sex/group were included in each group as potential replacement animals. However, these animals were not required for blood sampling, and were sacrificed and discarded at study termination. Levels of MK-0869 were quantified using

- **Other:** None.

Results:

- **Clinical signs:** There were no treatment-related clinical signs of toxicity.

- **Mortality:** There was no treatment-related mortality. Two death occurred during week 1 as a result of dosing accidents. Histopathological findings for both Female #99-5581 (3 doses) in the control group, that received 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water once per day, and Female #99-5631 (2 doses) that received MK-0869 Formulation M at 500 mg/kg/day were observed in the lungs and consisted of suppurative inflammation in the mediastinum.

- **Body weights:** Impairment of body weight gain was observed for male and female mice that received MK-0869 Formulation NB at 750 mg/kg B.I.D.

Body weights for male controls, which received 5% methylcellulose and 0.02% sodium lauryl sulfate in deionized water, at pretest and week 4 were 34.1 and 36.6 g, respectively. Body weights for corresponding female controls at pretest and week 4 were 24.5 and 27.1 g, respectively. Body weight gains for male and female mice that received MK-0869 Formulation M at 500 mg/kg/day were 113.7 and 100.4% of controls, respectively.

Body weights for male controls, which received 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water once per day, at pretest and week 4 were 33.3 and 36.6 g, respectively. Body weights for corresponding female controls at pretest and week 4 were 24.7 and 28.2 g, respectively. Body weight gains for male mice that received MK-0869 Formulation NB at 25, 500, 1000, 1250, and 1500 mg/kg/day were 84.3, 101.5, 65.2, 122.2, and 81.1% of the control, respectively. Body weight gains for female mice that received MK-0869 Formulation NB at 25, 500, 1000, 1250, and 1500 mg/kg/day were 71.5, 87.1, 73.2, 69.4, and 91.2% of the control, respectively.

Body weights for male controls, which received 3% hydroxypropyl cellulose, 15% sucrose, and 0.14% sodium lauryl sulfate in deionized water twice per day, at pretest and week 4 were 35.8 and 37.9 g, respectively. Body weights for corresponding female controls at pretest and week 4 were 24.9 and 27.5 g, respectively. Body weight gains for male mice that received MK-0869 Formulation NB at 12.5, 250, 500, 625, and 750 mg/kg B.I.D. were 122.2, 109.5, 83.4, 94.1, and 69.3% of the control, respectively. Body weight gains for female mice that received MK-0869 Formulation NB at 12.5, 250, 500, 625, and 750 mg/kg B.I.D. were 69.2, 95, 112.9, 63.9, and 30.2% of the control, respectively.

- **Food consumption:** Food consumption was suppressed >10% for female mice that received MK-0869 Formulation M at 500 mg/kg/day, MK-0869 Formulation NB at doses ≥25 mg/kg/day, and MK-0869 Formulation NB at a dose of 750 mg/kg B.I.D. Total mean food consumption for female mice that received MK-0869 Formulation M at a dose of 500 mg/kg/day was decreased to 89.8% of the control (4.9 g/day). Total mean food consumption for female mice that received MK-0869 Formulation NB at ≥25 mg/kg/day was decreased to 84.3-88.2% of the control (5.1 g/day), although, there was no dose response relationship. Total mean food consumption for female mice that received MK-0869 Formulation NB at 750 mg/kg B.I.D. was decreased to 90.9% of the control (4.4 g/day).

- **Ophthalmoscopy:** The sponsor reported that there were no treatment-related ophthalmic changes; however, no data was provided for independent verification.

- **Hematology:** There were no treatment-related hematological changes.

- **Clinical chemistry:** Cholesterol and triglyceride levels were generally elevated for MK-0869 groups; however, dose response relationships were weak and their relationship to treatment was unclear. Cholesterol and triglyceride levels for female mice that received MK-0869 Formulation M at 500 mg/kg/day were increased to 116.5 and 142.9% of controls (85 and 56 mg/dL), respectively. Cholesterol and triglyceride levels for male rats that received MK-0869 Formulation M at 500 mg/kg/day were increased to 111.3 and 136% of controls (133 and 75 mg/dL), respectively. Cholesterol levels for female mice that received MK-0869 Formulation NB at 25, 500, 1000, 1250, and 1500 mg/kg/day were increased to 113.5-124.7% of the control (89 mg/dL). Cholesterol levels for male mice that received MK-0869 Formulation NB at doses of 25, 1000, 1250, and 1500 mg/kg/day were increased to 120.7-127.6% of the control (116 mg/dL); however, no changes was observed at 500 mg/kg/day. Triglyceride levels for female mice that received MK-0869 Formulation NB at 25, 1000, 1250, and 1500 mg/kg/day were increased to 137.3-188.2% of the control (51 mg/dL). Triglyceride levels for male mice that received MK-0869 Formulation NB at doses of 25, 500, 1000, 1250, and 1500 mg/kg/day were increased to 132.4-196.9% of the control (65 mg/dL). Cholesterol levels for female mice that received MK-0869 Formulation NB at 250, 500, 625, and 750 mg/kg B.I.D. were increased to 115.7-133.7% of the control (89 mg/dL). Triglyceride levels for female mice that received MK-0869 Formulation NB at doses of 250, 500, 625, and 750 mg/kg B.I.D. were increased to 123.7 to 161% of the control (59 mg/dL). Cholesterol and triglyceride levels for male mice that received MK-0869 Formulation NB at doses from 12.5 to 750 mg/kg B.I.D. were not altered in a treatment-related manner.

- **Organ Weights:** Increases in absolute and relative liver weights were observed for MK-0869 treatment groups that appeared to correlate with histopathological findings of centrilobular hypertrophy in the liver. Absolute and relative liver weights were increased for male mice that received MK-0869 Formulation M at 500 mg/kg/day, male mice that received MK-0869 Formulation NB at doses ≥25 mg/kg/day, female mice that received MK-0869 Formulation NB at doses ≥500 mg/kg/day, and male and female mice that received MK-0869 at doses ≥12.5 mg/kg B.I.D. Dose response relationships for increased absolute and relative liver weights were not evident or weak.

Absolute and relative liver weights for male mice that received MK-0869 Formulation M at 500 mg/kg/day were increased to 110.85 and 107.3% of controls (1.4120 g and 4.25%), respectively.

Absolute liver weights for male mice that received MK-0869 Formulation NB at doses of 25, 500, 1000, 1250, and 1500 mg/kg/day were increased to 113.7-126.9% of the control (1.3212 g). Relative liver weights for male mice that received MK-0869 Formulation NB at doses of 25, 500, 1000, 1250, and 1500 mg/kg/day were increased to 107.7-127.4% of the control (4.01% B.W.). Absolute liver weights for female mice that received MK-0869 Formulation NB at doses of 500, 1000, 1250, and 1500 mg/kg/day were increased to 110.3-115.8% of the control (1.1025 g). Relative liver weights for female mice that received MK-0869 Formulation NB at doses of 500, 1000, 1250, and 1500 mg/kg/day were increased to 117.2, 121.2, 122.1, and 122.6% of the control (4.30% B.W.).

Absolute liver weights for male mice that received MK-0869 Formulation NB at 250, 500, 625, and 750 mg/kg B.I.D. were increased to 113.8-122.3% of the control (1.3803 g). Relative liver weights for male mice that received MK-0869 Formulation NB at 12.5, 250, 500, 625, and 750 mg/kg B.I.D. were increased to 110.2-136% of the control (4.03% B.W.). Absolute liver weights for female mice that received MK-0869 Formulation NB at 250, 500, 625, and 750 mg/kg B.I.D. were increased to 118.2-135.3% of the control (1.0332 g). Relative liver weights for male mice that received MK-0869 Formulation NB at 12.5, 250, 500, 625, and 750 mg/kg B.I.D. were increased to 107.7-134.8% of the control (4.17% B.W.).

- **Gross Pathology:** There were no treatment-related gross pathological findings.

- **Histopathology:** Histopathological findings were confined to the liver. Centrilobular hypertrophy was observed for male and female mice that received MK-0869 Formulation M at 500 mg/kg/day. For the S.I.D. regimen, centrilobular hypertrophy was observed for male mice that received MK-0869 Formulation NB at doses ≥25 mg/kg/day and female mice that received MK-0869 Formulation NB at doses ≥500 mg/kg/day. The incidence of cellular infiltration was increased for female mice that received MK-0869 Formulation NB at doses ≥25 mg/kg/day, although, there was no dose response relationship. For the B.I.D. regimen, centrilobular hypertrophy was observed for male mice that received MK-0869 Formulation NB at doses ≥12.5 mg/kg B.I.D. and female mice that received MK-0869 Formulation NB at doses ≥250 mg/kg B.I.D. The incidence of cellular infiltration was generally increased for female mice that received MK-0869 Formulation NB at doses ≥12.5 mg/kg B.I.D., although, there was no dose response relationship.

Histopathological findings for the liver in female and male mice that received MK-0869 Formulation M at doses of 0 and 500 mg/kg/day.

Liver	0 mg/kg/day		500 mg/kg/day	
	Female	Male	Female	Male
number examined	0	10	2	15
centrilobular hypertrophy	-	-	-	9
cellular infiltration	-	3	1	1

Histopathological findings for the liver in female and male mice that received MK-0869 Formulation NB at doses of 0, 25, 500, 1000, 1250, and 1500 mg/kg/day.

Liver	0		25		500		1000		1250		1500	
	F	M	F	M	F	M	F	M	F	M	F	M
number examined	10	10	15	15	15	15	15	15	15	15	15	15
centrilobular hypertrophy	1	4	1	11	14	12	12	15	15	14	13	15
cellular infiltration	5	0	9	1	12	1	9	0	10	1	7	1

Histopathological findings for the liver in female and male mice that received MK-0869 Formulation NB at doses of 0, 12.5, 250, 500, 625, and 750 mg/kg B.I.D.

Liver	0		12.5		250		500		625		750	
	F	M	F	M	F	M	F	M	F	M	F	M
number examined	10	10	15	15	15	15	15	15	15	15	15	15
centrilobular hypertrophy	1	1	1	4	10	15	15	13	15	15	13	15
cellular infiltration	7	3	10	0	7	4	11	5	11	2	9	4

- **Toxicokinetics:** Toxicokinetic analysis was confined to the parent compound, MK-0869. Plasma AUC levels for MK-0869 were greater in female mice than male mice (<2-fold). For male and female mice that received MK-0869 Formulation NB at doses of 25 to 1500 mg/kg/day or 12.5 to 750 mg/kg B.I.D., increases in AUC values were significantly less than proportional to dose. For male and female mice that received MK-0869 Formulation NB at doses of 25 to 1500 mg/kg/day, a plateau in systemic exposure to the parent compound was evident at doses ≥500 mg/kg/day. For male mice that received MK-0869 Formulation NB at doses of 12.5 to 750 mg/kg B.I.D., a plateau in systemic exposure to the parent compound was evident at doses ≥250 mg/kg B.I.D. For female mice that received MK-0869 Formulation NB at doses of 12.5 to 750 mg/kg B.I.D., a plateau in systemic exposure to the parent compound was evident at doses ≥500 mg/kg B.I.D. Corresponding total daily doses of MK-0869 Formulation NB using the S.I.D. or B.I.D. regimens produced similar AUC values. Administration of MK-0869 Formulation NB at doses up to 1500 mg/kg/day or 750 mg/kg B.I.D. produced systemic exposures to the parent compound that were generally ≤2-fold the systemic exposure to the parent compound obtained with MK-0869 Formulation M at 500 mg/kg/day. Absorption of MK-0869 Formulation M or NB was delayed, possibly due to the insolubility of the test article in the vehicle.

Plasma toxicokinetic parameters for MK-0869 in mice that received MK-0869 Formulation M at a dose of 500 mg/kg/day for 29-30 days.

Dose, mg/kg/day	$AUC_{0\text{-}24\ hr}$, µg·hr/mL		C_{max}, µg/mL		T_{max}, hr	
	Female	Male	Female	Male	Female	Male
500	28.8	21.1	3.24	2.71	4.0	8.0

Plasma toxicokinetic parameters for MK-0869 in mice that received MK-0869 Formulation NB at doses of 25, 500, 1000, 1250, and 1500 mg/kg/day for 29-30 days.

Dose, mg/kg/day	$AUC_{0\text{-}24\ hr}$, µg·hr/mL		C_{max}, µg/mL		T_{max}, hr	
	Female	Male	Female	Male	Female	Male
25	17.2	12.8	2.53	2.14	2.0	2.0
500	57.4	33.7	6.51	4.72	6.0	2.0
1000	59.2	39.3	7.32	4.69	6.0	2.0
1250	47.4	35.1	6.50	4.84	8.0	6.0
1500	68.3	37.9	8.42	5.62	6.0	2.0

Plasma toxicokinetic parameters for MK-0869 in mice that received MK-0869 Formulation NB at doses of 12.5, 250, 500, 625, and 750 mg/kg B.I.D. for 29-30 days.

Dose, mg/kg B.I.D.	$AUC_{0\text{-}24\ hr}$, µg·hr/mL		C_{max}, µg/mL		T_{max}, hr	
	Female	Male	Female	Male	Female	Male
12.5	14.4	12.9	2.19	2.20	8.0	8.0
250	51.8	43.7	5.61	4.90	8.0	8.0
500	62.5	39.3	8.95	4.42	8.0	8.0
625	68.7	39.4	6.77	5.01	8.0	8.0
750	60.1	44.6	7.24	5.72	8.0	8.0

Key Study Findings: The toxicology and toxicokinetic profiles of two MK-0869 formulations of different particle size, were assessed in CD-1 mice during a 5-week treatment period (28 to 29 days). MK-0869 Formulation M (average particle size, ———) was administered to mice at oral doses of 0 and 500 mg/kg/day. MK-0869 Formulation NB (average particle size, ———) was administered using S.I.D. and B.I.D. regimens. For the S.I.D. regimen, MK-0869 Formulation NB was administered at doses of 0, 25, 500, 1000, 1250, and 1500 mg/kg/day. For the B.I.D. regimen, MK-0869 Formulation NB was administered at doses of 0, 12.5, 250, 500, 625, and 750 mg/kg B.I.D. (total daily doses of 0, 25, 500, 1000, 1250, and 1500 mg/kg, respectively). Histopathological findings were confined to the liver. Centrilobular hypertrophy was observed for male and female mice that received MK-0869 Formulation M at 500 mg/kg/day. For the S.I.D. regimen, centrilobular hypertrophy was observed for male mice that received MK-0869 Formulation NB at doses ≥25 mg/kg/day and female mice that received MK-0869 Formulation NB at doses ≥500 mg/kg/day. For the B.I.D. regimen, centrilobular hypertrophy was observed for male mice that received MK-0869 Formulation NB at doses ≥12.5 mg/kg B.I.D. and female mice that received MK-0869 Formulation NB at doses ≥250 mg/kg B.I.D. Toxicokinetic analysis was confined to the parent compound, MK-0869. Plasma AUC levels for MK-0869 were greater in female mice than male mice (<2-fold). For male and female mice that received MK-0869 Formulation NB at doses of 25 to 1500 mg/kg/day or 12.5 to 750 mg/kg B.I.D., increases in AUC values were significantly less than proportional to dose. For male and female mice that received MK-0869 Formulation NB at doses of 25 to 1500 mg/kg/day, a plateau in systemic exposure to the parent compound was evident at doses ≥500 mg/kg/day. For male mice that received MK-0869 Formulation NB at doses of 12.5 to 750 mg/kg B.I.D., a plateau in systemic exposure to the parent compound was evident at doses ≥250 mg/kg B.I.D. For female mice that received MK-0869 Formulation NB at doses of 12.5 to 750 mg/kg B.I.D., a plateau in systemic exposure to the parent compound was evident at doses ≥500 mg/kg B.I.D. Corresponding total daily doses of MK-

0869 Formulation NB using the S.I.D. or B.I.D. regimens produced similar AUC values. Administration of MK-0869 Formulation NB at doses up to 1500 mg/kg/day or 750 mg/kg B.I.D. produced systemic exposures to the parent compound that were generally ≤2-fold the systemic exposure to the parent compound obtained with MK-0869 Formulation M at 500 mg/kg/day. Absorption of MK-0869 Formulation M or NB was delayed, possibly due to the insolubility of the test article in the vehicle.

Fourteen-Week Oral Range-Finding Study in Mice (Report Date/Number TT #96-046-0).

Testing Laboratory: Merck Institute for Therapeutic Research
Merck Research Laboratories
Merck & Co., Inc.
West Point, PA 19486

Study Started: July 8, 1996

Study Completed: March 4, 1997

GLP Compliance: Statements of Compliance with GLP Regulations and the Quality Assurance Unit were included.

Animals: Crl:CD-1®(ICR)BR mice were used in this study. At the start of treatment, animals were 36 days old and body weight ranges were 23.9-34.0 g for male mice and 16.1-25.7 g for female mice.
Drug Batch: L-754,030-004H, batch #002.

Methods: Mice received L-754,030 by oral gavage at doses of 0, 25, 125, 250, 500, and 1000 mg/kg/day for 14 weeks. This study was intended as dose range finding study for a carcinogenicity study with L-754,030 in mice. Mice in the control group received the vehicle, 0.5% aqueous methyl cellulose/0.02% sodium lauryl sulfate. There were 10 mice/sex/group. The dose volume was 10 mL/kg. Animals were observed daily for mortality and clinical signs of toxicity. Body weight was measured prior to the start of treatment, once during the first week of treatment, and twice per week thereafter. Food consumption was measured once a week. Ophthalmic examinations were performed at week 6 and 12 with all animals in the control and 1000 mg/kg/day groups. Blood for determination of hematological and biochemical parameters was collected at week 14, prior to necropsy. Complete necropsy examinations were performed on all animals in the study. Absolute and relative organ weights were determined for the brain, heart, testes, kidneys, liver, and spleen. Organ and tissues were collected, processed, and subjected to histopathological analysis for 5 mice/sex of the control and 1000 mg/kg/day groups as follows: salivary gland, esophagus, stomach, small intestine, large intestine, liver, gall bladder, pancreas, adrenal gland, pituitary gland, thyroid gland, parathyroid gland (when present in thyroidal sections), kidneys, urinary bladder, ovary/testis (to include epididymis), uterus, prostate, heart, lung, thymus, lymph node, spleen, skin from mammary region, mammary gland (when present in skin section), bone (with marrow and joint), skeletal muscle, brain (to include cerebral cortex, and subcortical white matter, thalamus and hypothalamus, mid-brain, cerebellum and pons, and

medulla), cervical spinal cord, peripheral nerve (sciatic), eye (to include optic nerve and Harder's gland). The kidneys, adrenal glands, and liver from the remaining 5 mice/sex of the control and 1000 mg/kg/day groups were examined. Additionally, all macroscopic and ophthalmic changes from all mice, livers from all male and female mice, and kidneys from all females were processed and subjected to microscopic analysis.

Results:

1. Observed Effects: The sponsor did not report any treatment-related observed effects.

2. Mortality: There were no treatment-related deaths. One female mouse in the 25 mg/ kg/day group and one male mouse in the 250 mg/kg/day group died during the treatment period. Both deaths were attributed to intubation accidents.

3. Body Weight and Food Consumption: There were no treatment-related effects on body weight gain or food consumption. Body weights for male control at weeks –1 and 13 were 28.0 and 37.8 g, respectively. Body weight gains for male mice that received
L-754,030 at 25, 125, 250, 500, and 1000 mg/kg/day were 109.3, 117.6, 113.9, 119.9, and 98.6% of the control, respectively. Body weights for female control at weeks –1 and 13 were 22.8 and 29.1 g, respectively. Body weight gains for female mice that received
L-754,030 at 25, 125, 250, 500, and 1000 mg/kg/day were 128.6, 117.5, 110.35, 127.2, and 101.9% of the control, respectively.

4. Hematology: There were no treatment-related changes of hematological parameters.

5. Blood Biochemistry and Urinalysis: A number of blood biochemical changes were observed; although, they appeared to have little toxicological significance. Blood levels of Na^+, K^+, and Cl^- were not determined for female mice that received L-754,030 at 1000 mg/ kg/day. Urinalysis parameters were not measured. Cholesterol and triglyceride levels for female mice that received L-754,030 at 1000 mg/kg/day were increased to 108.9 and 156.9% of the control (90 and 65 mg/mL), respectively. Triglyceride levels for male mice that received L-754,030 at 1000 mg/kg/day were increased to 147.8% of the control
(69 mg/mL). Aspartate transaminase activities for female mice that received L-754,030 at 25, 125, 250, 500, and 1000 mg/kg/day were decreased to 52, 59.8, 52, 86.3, and 52% of the control (102 U/L), respectively. Ca^{2+} levels for female mice that received L-754,030 at doses between 25 and 1000 mg/kg/day were elevated to 102-109% of the control (9.0 mg/ dL). Ca^{2+} levels for male mice that received L-754,030 at doses between 25 and 1000 mg/ kg/day were decreased to 95-97% of the control (9.9 mg/dL).

6. Ophthalmic Examination: The sponsor stated that there were no treatment-related ophthalmic effects; although, data was not presented for independent analysis.

7. Organ Weights: Absolute and relative liver weights were increased for male and female mice that received L-754,030. Increased liver weight correlated with histopathological findings for the liver of centrilobular hepatocellular hypertrophy. Absolute liver weights for male mice that received L-754,030 at 25, 125, 250, 500, and 1000 mg/ kg/day were increased to 111, 117.9, 114.4, 116.1, and

111.8% of the control (1.299 g), respectively. Relative liver weights for male mice that received L-754,030 at 25, 125, 250, 500, and 1000 mg/kg/day were increased to 106, 107.9, 104.2, 112.6, and 109.4% of the control (3.82%), respectively. Absolute liver weights for female mice that received L-754,030 at 25, 125, 250, 500, and 1000 mg/kg/day were increased to 108.6, 112.6, 112.3, 116.1, and 111.4% of the control (1.140 g), respectively. Relative liver weights for female mice that received L-754,030 at 25, 125, 250, 500, and 1000 mg/kg/day were increased to 103.6, 107.7, 114, 117.8, and 107.7% of the control (4.15%), respectively.

8. Gross Pathology: Gross pathological changes were not reported.

9. Histopathology: Centrilobular hepatocellular hypertrophy of the liver was observed for male mice at doses ≥25 mg/kg/day and for female mice at doses ≥125 mg/kg/day; however, it is considered to have little or no toxicological significance. Hydropic degeneration of tubules in the kidneys was observed for female mice at 1000 mg/kg/day.

Histopathological changes for mice that received L-754,030 by oral gavage at doses of 0, 25, 125, 250, 500, and 1000 mg/kg/day for 14 weeks.

Organ/Tissue	0		25		125		250		500		1000	
	F	M	F	M	F	M	F	M	F	M	F	M
Liver -centrilobular hepatocellular hypertrophy	0	0	0	7	3	8	1	5	5	8	1	8
Kidney -tubule hydropic degeneration	0	-	0	-	0	-	0	-	0	-	6	-

Mice received L-754,030 by oral gavage at doses of 0, 25, 125, 250, 500, and 1000 mg/kg/day for 14 weeks. This study was intended as dose range finding study for a carcinogenicity study with L-754,030 in mice. The dose of 500 mg/kg/day could be considered a tolerated dose. The target organs of toxicity were the liver and kidneys. Centrilobular hepatocellular hypertrophy of the liver was observed for male mice at doses ≥25 mg/kg/day and for female mice at doses ≥125 mg/kg/day; however, it was considered to have little or no toxicological significance. Hydropic degeneration of tubules in the kidneys was observed for female mice at 1000 mg/kg/day.

Dogs

1. 4-Week Intravenous Toxicity Study of L-758298 (Study TT #95-009-0)

Testing Laboratory: Merck Research Laboratories
Merck & Co., Inc.
West Point, PA

Compliance with Good Laboratory Practice and Quality Assurance Requirements: Sponsor provided statements of compliance.

Date Study Started: February 21, 1995

Date Study Completed: July 18, 1995

Animals: Male (body weight range of 7.5 to 14.2 kg; 43 to 56 weeks of age) and female (body weight range of 5.9 to 11.3 kg; 43 to 56 weeks of age) Beagle dogs.

Methods: Five groups of 8 dogs each (4 males and 4 females) were intravenously administered 0, 0.5, 2, 8 and 32 mg/kg/day of L-758,298, respectively, for 4 weeks via the cephalic vein. The basis for dose selection was not provided by the sponsor. Vehicle was 0.9% Sodium Chloride Injection, USP. Dosing volume was 3.2 ml/kg, not exceeding an injection rate of 40 ml/min.

Mortality and clinical signs of toxicity were observed daily. Body weight was measured once during pretest and once weekly during treatment. Food consumption was measured 3-4 days during each week of treatment.

Blood samples for hematology and blood chemistry examination were withdrawn via the jugular sinus from all dogs prior to study initiation and during Weeks 2 and 4 from all remaining dogs. Urine samples for urinalysis were collected from all dogs prior to study initiation and during Week 4 from all remaining dogs.

Ophthalmic examinations were performed on all dogs prior to study initiation and during Week 4 in all remaining dogs. Electrocardiograms were recorded from all dogs prior to study initiation and from all remaining dogs 3-6 hrs post-dose on one occasion during Week 3.

Blood samples for future determination of L-758,298 plasma levels were collected via the jugular vein on Day 1 at 0.033, 0.117, 1, 2, 5, 7, 10 and 24 hrs after dosing in all remaining animals, during Week 4 at 0.033, 0.117, 1, 2, 5, 7 and 10 hrs after dosing in all animals.

All dogs underwent complete necropsies at scheduled termination. (Details were not provided by the sponsor.) Organ weights of brain, pituitary, spleen, heart, kidney, liver, adrenal, thyroid, testis/ovary and prostates were determined for all animals.

Sections of the following tissues from all control and high-dose dogs were prepared by routine methods, stained with hematoxylin and eosin, and examined microscopically: injection site, salivary gland, stomach, small intestine, large intestine, liver, pancreas, adrenal, thyroid, parathyroid, pituitary, kidney, urinary bladder, ovary/testis, uterus/prostate, skin, mammary gland, lung, heart, spleen, lymph node, thymus, skeletal muscle, bone, bone marrow, brain, cervical spinal cord, sciatic nerve, eye and Harder's gland. Tissues with gross pathology were microscopically examined for all animals.

Results:

1. Observed Effects: The 8 and 32 mg/kg i.v. injections produced slight to marked swelling, firmness of the forelimb and reddish discoloration of the skin. Animals displayed decreased weight bearing on the affected limbs, painful limbs and non-palpable veins. In some cases, when i.v. injections could not be administered by the cephalic veins, i.v. injections were delivered by the saphenous veins.

Males and females in the 32 mg/kg group were removed from the study after a total of 3-4 and 1-2 doses, respectively.

2. Mortality: Males and females in the 8 mg/kg group were removed from the study after a total of 4-8 and 3-8 doses, respectively, and were sacrificed in moribund condition.

3. Body Weight: Mean body weights of control males were 11.7 and 11.8 kg during Weeks 1 and 4, respectively. Mean body weights of control females were 9.0 and 9.2 kg during Weeks 1 and 4, respectively. There were no treatment-related effects on mean body weights.

4. Food Consumption: Mean food consumption of control males was 335 and 342 g/day during Weeks 1 and 4, respectively. Mean food consumption of control females was 241 and 282 g/day during Weeks 1 and 4, respectively. There were no treatment-related effects on mean food consumption.

5. Hematology: When one compares the erythrocyte count (millon/mm^3), hemoglobin concentration (g/ml) and hematocrit (%) at pretest and prior to early sacrifice in dogs of the 8 mg/kg group, values were decreased by -9.25%, -9.5% and -11.0%, respectively. These changes may reflect intravascular hemolysis. There were no treatment-related hematological changes in the 0.5 and 2 mg/kg groups.

6. Blood Chemistry: There were no treatment-related effects on blood chemistry parameters.

7. Urinalysis: There were no treatment-related effects on urinalysis parameters.

8. Ophthalmic Examination/Electrocardiograms: There were no treatment-related ophthalmic effects. There were no treatment-related effects on ECGs.

9. Organ Weights: There were no treatment-related effects on organ weights.

10. Gross Pathology: The 8 and 32 mg/kg i.v. injections produced slight to marked swelling, firmness of the forelimb and reddish discoloration of the skin. These effects were not seen in either the 2 mg/kg/day group or the control group.

11. Histopathology: In the 32 mg/kg/day group, 4/4 males and 4/4 females had venous thrombosis, and 2/4 males and 2/4 females had venous vascularization; these effects were not seen in any other group.

12. Plasma Levels of Drug: Sponsor stated that data for plasma levels of L-758,298 will be provided at a future date.

In summary, the no effect i.v. dose of L-758,298 was 2 mg/kg/day in the dog. The 8 and 32 mg/kg/day doses produced slight to marked swelling, firmness of the forelimb and reddish discoloration of the skin. The 32 mg/kg/day i.v. dose produced venous thrombosis and vascularization. No target organs of toxicity were identified.

5-Week intravenous toxicity study in dogs (TT#95-009-0)

This study has previously been reviewed under IND [redacted] original submission dated September 28, 1995 (pharmacologist's review of IND [redacted] dated April 15, 1996). In this amendment, the sponsor submitted the toxicokinetic data for this study, which is reviewed below.

Toxicokinetics: In this study, animals were treated with L-758, 298 at 0.50, 2.0, 8.0, and 32.0 mg/kg i.v. doses. The plasma concentrations were studied at 0.5 and 2.0 mg/kg. On Day 1, animals were bled at 2 min, 7 min, 1, 2, 5, 7, 10, and 24 hours post-dose. At week 4, samples were collected at 0, 2, and 7 min, and at 1, 2, 5, 7 and 10 hours post-dose. Plasma concentrations of pro- and active drug were determined by [redacted] method. Maximum mean plasma concentrations of the active drug, L-754, 030, were attained very rapidly (within 2 minutes post-dose). The systemic exposure or AUC_{0-24h} increased in a slightly greater than dose-proportional manner. There were no gender-related differences in toxicokinetics at 0.5 or 2.0 mg/kg doses. The following table (from vol. 2, pg. 256 and 257 of sponsor's submission) summarizes the mean toxicokinetic parameters of L-754, 030 after i.v. administration of L-758, 298 at 0.5 and 2.0 mg/kg, i.v. in dogs.