PILOT RAT TERATOLOGY STUDY

T-3352

FINAL REPORT

HAZLETON
LABORATORIES AMERICA, INC.
9200 LEESBURG TURNPIKE, VIENNA, VIRGINIA 22180, U.S.A.
PHONE (703) 893-5400, TELEX 899436 (HAZLABS VINA), CABLE HAZLABS WASH DC

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Submitted to

3M Company
St. Paul, Minnesota

May 11, 1983

HAZLETON
LABORATORIES AMERICA, INC.
9200 LEESBURG TURNPIKE, VIENNA, VIRGINIA 22180, U S A

SUBJECT: Pilot Rat Teratology Study
Project No. 154-159

We, the undersigned, hereby declare that the work was performed under our supervision, according to the procedures herein described.

Study Director:

LAWRENCE T. WETZEL, Ph.D.
Staff Scientist
Department of Toxicology

Laboratory Supervision:

GEORGE A. BURDOCK, Ph.D.
Rodent Toxicology I
Department of Toxicology

Teratology:

RUTH S. DURLOO, B.S.
Research Associate
Department of Toxicology

Report Preparation:

MARCIA A. MENSE, B.A.
Technical Writer
Scientific Resources Department

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HAZLETON LABORATORIES AMERICA, INC.
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SPONSOR: 3M Company

DATE: May 11, 1983

MATERIAL: T-3352

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Pilot Rat Teratology Study
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SUMMARY

T-3352, suspended in corn oil at concentrations intended to deliver 1, 10, 37.5, or 75 mg/kg/day (Groups 2, 3, 4, and 5, respectively), was administered by oral intubation to four groups of mated female Sprague-Dawley S-D® rats on gestation Days 6 through 15. A fifth group (Group 1) of mated females served as the control and received the vehicle only.

Apparent compound-related deaths occurred in seven of seven Group 5 and three of seven Group 4 females on or before Day 17 of gestation. Clinical observations in Groups 4 and 5 included hunched, thin appearance, languid behavior, and apparent anorexia. Body weight gain during treatment was less in Group 3, 4, and 5 females compared to control values obtained during that time. The terminal body weight minus the gravid uterine weight was decreased in Group 4 females compared to control. A number of gross pathology findings at necropsy or sacrifice were noted in Groups 4 and 5 and these included incidences of liver, adrenal, lung, and/or gastrointestinal tract effects.

Evaluation of mean fetal weight data indicated that fetal weights of both sexes decreased with increasing dose level. A significant decrease in mean fetal weight occurred in Group 4 males and females, although data for only two litters were available for that group. Cleft palate was found in two of six Group 4 fetuses (one fetus in each litter) examined by Wilson's technique. Incompletely descended testes were noted in one fetus from each Group 4 litter evaluated for visceral defects. Incidences of skeletal variants were similar for all groups.

INTRODUCTION

This study was designed to determine the maternal and embryo/fetal toxicity of T-3352 when administered by gavage to pregnant rats during the period of fetal organogenesis for the purpose of setting dose levels for an expanded teratology study. The rats were placed in breeding on January 17, 1983, and cesarean sections were completed on February 18, 1983. This report presents the methods and results from this study.

CONTROL AND TEST MATERIALS

The vehicle and control material, Duke's® Corn Oil (C. F. Sauer Co., Richmond, Virginia), lot 80235, was received on January 24, 1983, and was stored at room temperature.

The test material, T-3352, an off-white solid, was received from the sponsor on January 5, 1983, and was stored at room temperature. The test material was assumed to be 100% active compound. Information on methods of synthesis and stability, as well as data on composition or other characteristics which define the test material, are on file with the sponsor.

TEST ANIMALS

Sexually mature cesarean-derived Sprague-Dawley Crl:CD® (SD)BR rats were chosen for this study because they are sensitive to a number of agents which are known to be embryotoxic and/or teratogenic. Rats have historically been used in safety evaluation studies of this type and are required by the regulatory agencies. Thirty-six twelve to fourteen-week old male rats and thirty-six eight to ten-week old female rats were received from Charles River Breeding Laboratories, Kingston, New York, on December 22, 1982. The rats were held in quarantine for three and one-half weeks, during which time a health status examination was performed by a staff veterinarian.

The rats were housed one male and one female per cage during breeding. Following confirmation of mating, the females were housed individually in elevated wire-mesh cages with food (Purina Rodent Laboratory Chow® 5001) and tap water available ad libitum. The females were uniquely identified by ear tag after mating was confirmed. Temperatures in the study room ranged from 70 to 78°F with a relative humidity between 44 and 69%.

METHODS

Mating Period

During the mating period, one female was paired with one male until mating was confirmed or until two weeks had elapsed. Daily vaginal

examinations of each female were performed to detect the presence and viability of sperm or the presence of a copulatory plug. The day of observation of sperm or copulatory plug was designated as "Day 0" of gestation.

Groups and Dosage Levels

Upon confirmation of mating, each female was assigned to one of the following groups.

Group	Dose[a] mg/kg/day	Number of Females
1	0	6
2	1	6[b]
3	10	7
4	37.5	7
5	75	7

[a] Based on individual animal body weights at each weighing interval during the dosing period.

[b] An additonal Group 2 female was confirmed to have mated on a day when no other matings were confirmed. Because this animal's dose level would have been known at cesarean sacrifice, she was removed from the study.

The females were placed into the dose groups one at a time beginning with the high-dose group and continuing sequentially through the control group until all mated females were assigned to a group.

All males were sacrificed via carbon dioxide asphyxiation and discarded without necropsy once a sufficient number of matings were confirmed.

Compound Preparation and Administration

Suspensions of the test material in the corn oil vehicle were prepared on a weight per volume basis. The required amount of compound for levels 2, 3, or 4 was weighed into a homogenizer on an electronic Arbor 126 balance. Compound for Group 5 was weighed on powder paper on an Arbor 126 balance and rinsed into a Waring blender. Approximately 4 ml of corn oil was added and the test material was ground into a fine suspension. This suspension was rinsed from the homogenizer or the blender into a beaker and additional corn oil was added to produce the desired concentration. The suspension was mixed on a stirrer for approximately fifteen minutes. Prepared suspensions were mixed with a magnetic stirrer during dosing.

Females were given the appropriate dosing suspension or vehicle by oral intubation on a daily basis beginning on Day 6 and continuing through Day 15 of gestation. The test material was administered orally because of the relative ease and accuracy of dosing.

Fresh dosing suspensions were prepared weekly and stored under refrigeration (approximately $41^{o}F$). Samples of each test mixture as well as the vehicle were sent to the sponsor for analysis.

Maternal Observations and Records

All animals were examined twice daily for mortality and moribundity (from day of receipt through study termination) and once daily for clinical signs of toxicity and pharmacologic effects (throughout

gestation). Body weights and food consumption were recorded on Days 0, 6, 8, 12, 16, and 20 of gestation.

Cesarean Sacrifice and Tissue Preservation

A gross necropsy was performed on all animals (found deads, moribund sacrifices, and terminal sacrifices). The uterus and ovaries from found dead animals and animals sacrificed because of moribundity were removed and examined for the number of implantation sites and corpora lutea, respectively. Prior to gestation Day 20, surviving females were assigned random numbers and all personnel performing cesarean sections and/or external, visceral, or skeletal examinations of the fetuses were unaware of the dose level from which the animals were derived. On Day 20 of gestation, all surviving females were weighed and sacrificed by carbon dioxide asphyxiation.

The uterus from each female was weighed and examined for the number and placement of uterine implantation sites, number of live and dead fetuses, early and late resorbing fetuses, and any abnormalities. The uterus of each animal was reweighed after the contents were removed. The ovaries were examined for the number of corpora lutea. Each live fetus was sexed, weighed, and examined for external abnormalities. Findings were recorded.

Beginning at the ovarian end of the right uterine horn, the first six fetuses, regardless of sex, were selected for further evaluation. After the external examination was completed, the first,

third, and fifth fetuses were identified with a tag and fixed in Bouin's solution for soft tissue evaluation (Wilson, et al, 1965). The second, fourth, and sixth fetuses were eviscerated, tagged, and processed for skeletal examination using a technique modified from that reported by Staples (Staples and Schnell, 1964).

Statistical Analyses

Mean maternal body weight gains (Days 6-16 and 0-20), total food consumption, gravid uterine weight, terminal body weight minus gravid uterine weight, resorption incidence, percent males, and fetal viability were analyzed in the following order. Levene's test for homogeneity of variances (Levene, 1960; Draper and Hunter, 1969) was performed and if the variances proved to be homogeneous, the data were analyzed by one-way classification analysis of variance (ANOVA) (Winer, 1971). If the variances proved to be heterogeneous, a series of transformations was performed until variance homogeneity was achieved. These transformations were: $\log_{10}$, square (X^2), square root ($X^{\frac{1}{2}}$), reciprocal (1/X), angular (arcsine $X^{\frac{1}{2}}$), and rank, in that order. If rank transformation was ineffective in removing variance heterogeneity, ANOVA of ranked data was completed. If ANOVA of untransformed or transformed data was significant, Dunnett's t-test (Dunnett, 1955 and 1964) was used for control vs. compound-treated group mean comparisons. If ANOVA was not significant, the analysis was complete.

In addition to the above data, analysis of covariance (ANCOVA) (Winer, 1971) was used to analyze mean fetal weights. The litter was used as the experimental unit.

Levene's tests and ANOVA were evaluated at the 5.0% one-tailed probability level. Control vs. compound-treated group mean comparisons were evaluated at the 5.0% two-tailed probability level.

Statistical references are appended to this report, and statistically significant differences, as indicated by the aforementioned tests, are designated throughout this report by the term "significant" and/or as follows:

S+ = Significantly higher than the control value.

S- = Significantly lower than the control value.

Specimen, Raw Data, and Final Report Storage

All specimens, raw data, and the final report are stored in the archives of Hazleton Laboratories America, Inc.

RESULTS - MATERNAL DATA

Clinical Signs

Summary clinical signs are presented in Table 1.

Treatment-related clinical observations were noted in the Groups 4 and 5 animals during treatment and in Group 4 animals posttreatment. No Group 5 rats survived the treatment phase. Clinical signs for Groups 4 and 5 animals included hunched and thin appearance, languid behavior, urine stains, and bloody crusted eyes, eyelids, nose, mouth, legs, paws, or genitals. Anorexia, ataxia, dyspnea, rough coat and pale appearance were also noted for several Group 4 or 5 rats. One Group 4 rat aborted thirteen fetuses on Day 20 prior to sacrifice. Alopecia was noted for some rats in each dose group and the control group.

Mortality

All Group 5 females were found dead on or before Day 16 of gestation and three Group 4 females were found dead on or before Day 17 of gestation. All other animals survived to cesarean sacrifice on Day 20.

Body Weights

Individual and mean body weights and body weight changes are presented in Appendix 1. Mean weight values and body weight changes are presented in Table 2.

During gestation, lower than control mean body weight values were noted in Group 5 beginning on Day 8 and in Group 4 beginning on Day 12. Decreased body weight gain during gestation was noted in Groups 3

and 4. The decrease in the Group 4 animals was significantly different than the control value. Because of high mortality, Group 5 data were not included in statistical evaluation.

Food Consumption

Individual and mean food consumption values are presented in Appendix 2. Mean food consumption values are presented in Table 3.

Slightly decreased food consumption values were noted for Groups 4 and 5 compared to control on and after Day 8 of gestation. Mean total food consumption values for Groups 2, 3, and 4 were statistically comparable to control. Total food consumption for Group 5 was not determined because of mortality.

Gross Pathology

Summary gross pathology findings are presented in Table 4.

No gross lesions were noted at necropsy for animals from the control group or dose Groups 2 and 3. Findings noted for Group 4 animals were discolored liver, kidney, and adrenals; enlarged adrenals; dilated renal blood vessels, and red vaginal discharge. Findings noted for Group 5 animals were reddened and/or distended lungs, enlarged and/or friable liver, enlarged adrenals, discolored and/or thin glandular gastric mucosa, fluid in stomach or intestines, and red vaginal discharge.

Uterine Weights

Individual and mean uterine weights are presented in Appendix 3. Summary uterine weights are presented in Table 5.

Mean terminal body weights were comparable for Groups 1, 2, and 3, but the mean terminal body weight of the Group 4 females was decreased compared to control animal weights. There was an approximate 9% decrease in the mean gravid uterine weights of the Group 2, 3, and 4 animals compared to the control mean weight. For Group 2, this may be attributable to one dam with eight early resorbing fetuses. Terminal body weight minus gravid uterine weights were comparable for Groups 1, 2, and 3, but that weight was decreased in the Group 4 animals compared to the control value.

RESULTS - CESAREAN DATA

Individual and mean ovarian, uterine, and litter data are presented in Appendix 4. Individual and mean live fetal data are presented in Appendix 5. Summary ovarian, uterine, and litter data are presented in Table 6.

Pregnancy Rates, Corpora Lutea, Uterine Implantations, and Implantation Efficiency

Pregnancy rates were 100 percent for Groups 1, 2, and 3, and 85.7 percent for Groups 4 and 5. The mean number of corpora lutea and the mean number of uterine implantations were comparable for all groups as were mean implantation efficiencies.

Fetal Viability, Weight, and Sex

Mean resorption incidences were higher for Groups 2 and 4 than for the control group, but these differences were not statistically significant. The increased incidence in Group 2 may be attributable to one dam with eight resorbing fetuses. Mean incidence of fetal viability and percent males were comparable for Groups 1-4. Mean live fetal weights of both sexes decreased with increasing dose level with Group 4 values significantly lower than control.

Fetal Development

Individual visceral findings are presented in Appendix 6; individual skeletal findings are presented in Appendix 7. Mean incidence values for visceral and skeletal findings are presented in Table 7 (based on number of fetuses) and Table 8 (based on number of litters).

Gross external examination of fetuses at cesarean section revealed one abnormal fetus from a Group 3 dam. This fetus had no tail and appeared to have no thoracic or lumbar vertebrae. However, no skeletal evaluation was done on this fetus so that finding could not be verified.

Skeletal examination of the selected fetuses revealed that the incidence of skeletal variants was comparable for the control group and treated groups. No skeletal anomalies were noted. The incidence of visceral variants was markedly higher for Group 4 when compared to the

control group (50 percent vs. 5.56 percent). However, only two Group 4 litters were available for evaluation compared to six of the control group. Variants noted were dilated renal pelves (Group 2), dilated ureters (Groups 1 and 4), and incompletely descended testes (Group 4).

Cleft palate was found in two of six Group 4 fetuses examined by Wilson sectioning. That anomaly was noted in one fetus from each of the two litters from which the fetuses selected for visceral examination were derived. No other visceral anomalies were observed.

Table 1
Clinical Observations During Gestation
Pilot Rat Teratology Study of T-3352

OBSERVATION	Group 1 - 0 mg/kg Day of Gestation			Group 2 - 1 mg/kg Day of Gestation			Group 3 - 10 mg/kg Day of Gestation			Group 4 - 37.5 mg/kg Day of Gestation			Group 5 - 75 mg/kg Day of Gestation		
	0-5	6-15	16-20	0-5	6-15	16-20	0-5	6-15	16-20	0-5	6-15	16-20	0-5	6-15	16-20
Number of animals observed	6	6	6	6	6	6	7	7	7	6(1)	6(1)	4(1)	6(1)	6(1)	0
Hunched											5(1)	3(1)		6(1)	
Thin											2	3		6(1)	
Languid												3(1)		6(1)	
Anorexia											1	1		3	
Ataxia														2(1)	
Dyspnea												2			
Urine stains											3	3		4(1)	
Alopecia (paws, legs, chest, neck, hips, side, abdomen)	2	4	4	1	2	2		1	2	1	2			1	
Rough haircoat												1		1	
Bloody crust (nose, mouth, paws, eyelids, legs, genitals, eyes)											5(1)	3(1)		6(1)	
Pale												1			
Aborted fetuses/red vaginal discharge												1			

() = Nonpregnant female.

Table 2
Mean Maternal Body Weights and Body Weight Change
Pilot Rat Teratology Study of T-3352

Group and Dose Level		Body Weight (grams) Day of Gestation 0	6	8	12	16	20	Body Weight Change (±g) Days of Gestation 6-16	0-20[a]
1	Mean	231.60	263.57	264.17	285.23	304.07	366.52	+40.50	+134.920
0 mg/kg	S.D.	6.935	6.342	6.735	3.885	8.703	8.422	6.111	7.161
	N	6	6	6	6	6	6	6	6
2	Mean	239.38	259.25	266.23	283.70	304.90	363.95	+45.65	+124.57
1 mg/kg	S.D.	18.051	29.056	22.061	20.883	23.384	31.752	21.052	20.838
	N	6	6	6	6	6	6	6	6
3	Mean	240.93	271.81	267.73	280.87	301.56	359.67	+29.74	+118.74
10 mg/kg	S.D.	10.348	12.006	17.535	14.384	17.119	19.150	10.317	18.776
	N	7	7	7	7	7	7	7	7
4	Mean	250.25	263.63	265.10	245.65	227.98	317.00	-35.65[S-]	+60.70[S-]
37.5 mg/kg	S.D.	17.535	21.958	7.558	16.890	32.551	58.690	21.324	32.527
	N	6	6	6	6	4	2	4	2
5	Mean	238.90	259.47	246.37	206.80	-	-	-80.7	-
75 mg/kg	S.D.	18.020	13.286	9.816	17.331	-	-	-	-
	N	6	6	6	6	0	0	1	0

[a] Statistical group comparisons performed on rank-transformed data.

Table 3
Mean Food Consumption Values
Pilot Rat Teratology Study of T-3352

Group and Dose Level (mg/kg)		Food Consumption (gm/day) Day of Gestation 6	8	12	16	20	Total Food Consumption(g) Days 0-20[a]
1	Mean	18.28	19.22	19.92	19.97	26.13	430.23
0 mg/kg	S.D.	1.981	1.933	1.685	1.039	1.895	17.578
	N	6	6	6	6	6	6
2	Mean	18.32	19.32	20.05	23.00	26.72	445.45
1 mg/kg	S.D.	4.058	3.370	2.665	4.785	3.996	58.101
	N	6	6	6	6	6	6
3	Mean	17.20	14.80	19.14	17.71	24.26	394.17
10 mg/kg	S.D.	1.681	4.246	2.539	3.060	0.860	31.539
	N	7	7	7	7	7	7
4	Mean	18.90	12.20	15.73	16.75	15.27	411.43
37.5 mg/kg	S.D.	2.346	3.774	19.816	24.530	13.201	156.013
	N	6	6	6	4	3	3
5	Mean	20.03	9.62	8.55	-	-	-
75 mg/kg	S.D.	7.684	2.690	11.886	-	-	-
	N	6	6	6	0	0	0

[a] Statistical group comparisons performed on rank-transformed data.

Table 4
Summary of Gross Pathology Findings
Pilot Rat Teratology Study of T-3352

ORGAN AND DESCRIPTION	Group: 1	2	3	4	5
	Dose Level (mg/kg): 0	1	10	37.5	75
Number of rats examined	6	6	7	6(1)	6(1)
Number with no gross lesions	6	6	7	2(1)	0
LUNGS					
Reddened					2
Distended					1
LIVER					
Enlarged					2(1)
Yellow				3	
Friable consistency					1
KIDNEYS					
Greenish				1	
Blood vessels - dilated				1	
ADRENALS					
Enlarged				2	2(1)
Brown				1	
STOMACH					
Contained dark red fluid					1
Contained black fluid					(1)
Glandular mucosa - reddened					3(1)
- thin					1
- black focal areas					1

() = Nonpregnant female.

Table 4 - Continued
Summary of Gross Pathology Findings
Pilot Rat Teratology Study of T-3352

ORGAN AND DESCRIPTION	Group: 1	2	3	4	5
	Dose Level (mg/kg): 0	1	10	37.5	75
INTESTINES					
Filled with dark fluid material					1
OVARIES					
Autolyzed					1
VAGINA					
Red/bloody discharge/dark red fluid				3	2

Table 5
Mean Terminal Body Weights, Gravid Uterine Weights, and
Terminal Body Weights Minus Gravid Uterine Weights
Pilot Rat Teratology Study of T-3352

Group and Dose Level mg/kg		Terminal Body Weight grams	Gravid Uterine Weight grams	Terminal Body Weight Minus Gravid Uterine Weight[a] grams
1	Mean	366.52	77.30	289.22
0 mg/kg	S.D.	8.422	7.925	2.378
	N	6	6	6
2	Mean	363.95	68.33	295.62
1 mg/kg	S.D.	31.752	21.509	22.459
	N	6	6	6
3	Mean	359.67	68.34	291.33
10 mg/kg	S.D.	19.150	11.081	22.396
	N	7	7	7
4	Mean	317.00	67.40	249.60
37.5 mg/kg	S.D.	58.690	19.233	39.457
	N	2	2	2
5	Mean	-	-	-
20 mg/kg	S.D.	-	-	-
	N	0	0	0

[a] Statistical group comparisons performed on rank-transformed data.

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Key to Table 6

Pregnancy Rate (percent) = (number of pregnant rats/number of rats mated) x 100.

Survival Rate (percent) = (number of rats surviving to Day 20/number of rats placed on study) x 100.

Mean Implantation Efficiency (percent) = Group mean of ([implantations per litter/corpora lutea per litter] x 100).

Mean Resorption Incidence (percent) = Group mean of ([resorptions per litter/implantations per litter] x 100).

Mean Incidence of Fetal Mortality (percent) = Group mean of ([dead fetuses per litter/implantations per litter] x 100).

Mean Incidence of Fetal Viability (percent) = Group mean of ([live fetuses per litter/implantations per litter] x 100).

Table 6
Summary of Ovarian, Uterine, and Litter Data
Pilot Rat Teratology Study of T-3352

Group:	1	2	3	4	5
Dose Level (mg/kg):	0	1	10	37.5	75
Number of females with mating confirmed	6	6[a]	7	7	7
Number of females pregnant	6	6	7	6	7
Pregnancy rate (%)	100	100	100	85.7	85.7
Number of females surviving to Day 20	6	6	7	4	0
Survival rate (%)	100	100	100	57.1	0
Mean number of:					
Corpora lutea	15.0	16.8	15.6	16.2	15.7
Implantations	14.5	13.3	13.7	14.5	14.0
Resorptions	1.2	2.0	1.1	2.8	-
Fetuses - dead	0.0	0.0	0.0	0.0	-
- live	13.2	11.3	12.6	13.5	-
Indices calculated on a per litter basis:					
Mean implantation efficiency (%)	95.28	81.82	88.01	89.00	90.76
Mean resorption incidence (%)	7.95	15.75	7.66	13.25	-
Mean incidence of fetal mortality (%)	0.0	0.0	0.0	0.0	-
Mean incidence of fetal viability (%)	90.78	84.25	92.36	86.75	-
Live fetuses					
Mean body weight (g)					
Males - unadjusted	3.42	3.33	3.26	2.75	-
- covariance adjusted	3.40	3.37	3.27	2.72S-	-
Females - unadjusted	3.25	3.23	3.11	2.60	-
- covariance adjusted	3.25	3.22	3.12	2.60S-	-
Percent males	55.03	53.05	51.31	55.40	-

[a] An additional Group 2 female was confirmed to have mated on a day when no other matings were confirmed. Because this animal's dose level would have been known at cesarean sacrifice, she was removed from the study.

Table 7
Mean Incidence Values for Visceral and Skeletal Findings
Pilot Rat Teratology Study of T-3352

Group:	1	2	3	4
Dose Level (mg/kg):	0	1	10	37.5
Visceral				
Number of fetuses examined	18	18	21	6
Number of fetuses within normal limits	17	17	21	1
Number/percent of fetuses having one or more visceral variants	1/5.56	1/5.56	0	3/50.00
Number/percent of fetuses with:				
Dilated renal pelvis	0	1/5.56	0	0
Dilated ureter(s)	1/5.56	0	0	1/16.67
Incompletely descended testis(es)	0	0	0	2/33.33
Number/percent of fetuses having one or more visceral anomalies	0	0	0	2/33.33
Number/percent of fetuses with:				
Cleft palate	0	0	0	2/33.33
Skeletal				
Number of fetuses examined	18	17	21	6
Number of fetuses within normal limits	5	6	10	1
Number/percent of fetuses having one or more skeletal variants	13/72.22	11/64.71	11/52.38	5/83.33
Number/percent of fetuses with:				
Lagging Ossification				
Skull - incomplete closure (less than 75%)	4/22.22	2/11.76	1/4.76	0
- interparietal ossification incomplete	8/44.44	2/11.76	6/28.57	2/33.33
- supraoccipital ossification incomplete	9/50.00	7/41.18	6/28.57	3/50.00
- supraoccipital nonossification	0	0	0	0
- hyoid ossification incomplete	1/5.56	0	0	0
- hyoid - nonossified	9/50.00	2/11.76	3/14.29	0
- parietals irregularly ossified	4/22.22	3/17.65	5/23.81	0

Table 7 - Continued
Mean Incidence Values for Visceral and Skeletal Findings
Pilot Rat Teratology Study of T-3352

Group:	1	2	3	4
Dose Level (mg/kg):	0	1	10	37.5
Skeletal (Continued)				
Number of fetuses examined				
Number/percent having one or more skeletal variants	13	11	11	5
Number/percent of fetuses with:				
Lagging Ossification				
Rib-cage - sternebrae bipartite	0	1/5.88	2/9.52	4/66.67
- rib irregularly ossified	1/5.56	1/5.88		0
Vertebral column - thoracic centra ossification incomplete	0	0	1/4.76	0
- thoracic centra nonfused	1/5.56	1/5.88	1/4.76	1/16.67
- caudals less than three ossified	1/5.56	0	1/4.76	1/16.67
Bone alignment				
Extra rib(s)	1/5.56	0	1/4.76	0
Angulated rubs	2/11.11	1/5.88	0	0
Number/percent having one or more skeletal anomalies	0	0	0	0

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Key to Table 8

Mean Incidence of Visceral Anomalies (percent) = Group mean of ([number of fetuses with anomalies per litter/number of fetuses examined viscerally per litter] x 100).

Mean Incidence of Visceral Variants (percent) = Group mean of ([number of fetuses with variants per litter/number of fetuses examined viscerally per litter] x 100).

Mean Incidence of Skeletal Anomalies (percent) = Group mean of ([number of fetuses with anomalies per litter/number of fetuses examined skeletally per litter] x 100).

Mean Incidence of Skeletal Variants (percent) = Group mean of ([number of fetuses with variants per litter/number of fetuses examined skeletally per litter] x 100).

Table 8
Mean Incidence of Visceral and Skeletal Findings per Litter
Pilot Teratology Study of T-3352 in Rats

Group:	1	2	3	4
Dose Level (mg/kg):	0	1	10	37.5
Visceral Examination				
Number of litters examined	6	6	7	2
Number of litters with anomalous fetuses	0	0	0	2
Percent of litters with anomalous fetuses (%)	0	0	0	100.00
Number of litters with variant fetuses	1	1	0	2
Percent of litters with variant fetuses (%)	16.67	16.67	0	100.00
Mean values calculated on a per litter basis:				
Number of fetuses with variants	0.17	0.17	0	1.50
Incidence of variants(%)	5.56	5.56	0	50.00
Number of fetuses with anomalies	0	0	0	1.00
Incidence of anomalies (%)	0	0	0	33.33
Skeletal Examination				
Number of litters examined	6	6	7	2
Number of litters with anomalous fetuses	0	0	0	0
Percent of litters with anomalous fetuses (%)	0	0	0	0
Number of litters with variant fetuses	6	5	7	2
Percent of litters with variant fetuses (%)	100.00	83.33	100.00	100.00
Mean values calculated on a per litter basis:				
Number of fetuses with variants	2.17	1.83	1.57	2.50
Incidence of variants (%)	72.22	61.11	52.38	83.33
Number of fetuses with anomalies	0	0	0	0
Incidence of anomalies (%)	0	0	0	0

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Key to Appendix 1
Individual and Mean Maternal Body Weights
Pilot Rat Teratology Study of T-3352

P = Pregnant
NP = Not pregnant
C = Cesarean section performed (preceded by day of gestation)
FD = Found dead (preceded by day of gestation
A = Aborted pups (preceded by day of gestation)

[a] Body weight on day of death.
[b] Statistical group comparison performed on rank-transformed data.

Note: Body weights in parentheses are excluded from mean calculations and statistical evaluation.

Appendix 1
Individual and Mean Maternal Body Weights and Weight Changes
Pilot Rat Teratology Study of T-3352

Maternal Number	Date of Mating	Disposition	Body Weight (grams) - Day of Gestation						Weight Change (grams)	
			0	6	8	12	16	20	Days 6-16	Days 0-20[b]
			Group 1 - 0 mg/kg							
21606	1/18/83	P,20-C	229.2	271.1	272.3	288.8	309.9	367.2	38.8	138.0
21607	1/18/83	P,20-C	228.6	263.4	256.2	280.6	293.9	366.3	30.5	137.7
21608	1/19/83	P,20-C	235.4	265.7	269.8	289.7	313.6	374.7	47.9	139.3
21609	1/19/83	P,20-C	238.5	262.4	261.9	285.0	308.2	376.8	45.8	138.3
21610	1/19/83	P,20-C	237.6	266.6	267.6	286.5	306.1	358.1	39.5	120.5
21611	1/20/83	P,20-C	220.3	252.2	257.2	280.8	292.7	356.0	40.5	135.7
Mean			231.60	263.57	264.17	285.23	304.07	366.52	40.50	134.92
S.D.			6.935	6.342	6.735	3.885	8.703	8.422	6.111	7.161
			Group 2 - 1 mg/kg							
21613	1/18/83	P,20-C	238.6	216.6	257.6	284.5	300.5	336.4	83.9	97.8
21614	1/18/83	P,20-C	261.8	297.6	294.0	304.3	329.2	403.8	31.6	142.0
21615	1/19/83	P,20-C	250.7	285.3	290.4	309.4	333.8	400.9	48.5	150.2
21616	1/19/83	P,20-C	213.6	251.8	238.4	253.9	274.7	348.6	22.9	135.0
21617	1/19/83	P,20-C	223.4	245.2	251.1	269.7	285.5	330.9	40.3	107.5
21618	1/20/83	P,20-C	248.2	259.0	265.9	280.4	305.7	363.1	46.7	114.9
Mean			239.38	259.25	266.23	283.70	304.90	363.95	45.65	124.57
S.D			18.051	29.056	22.061	20.883	23.384	31.752	21.052	20.838

Appendix 1 - Continued
Individual and Mean Maternal Body Weights and Weight Changes
Pilot Rat Teratology Study of T-3352

Maternal Number	Date of Mating	Disposition	Body Weight (grams) - Day of Gestation						Weight Change (grams)	
			0	6	8	12	16	20	Days 6-16	Days 0-20[b]
			Group 3 - 10 mg/kg							
21620	1/18/83	P,20-C	241.5	264.3	257.6	274.9	280.9	344.7	16.6	103.2
21621	1/18/83	P,20-C	242.1	271.6	265.9	278.1	301.5	359.3	29.9	117.2
21622	1/19/83	P,20-C	226.9	256.9	245.2	267.3	300.5	360.5	43.6	133.6
21623	1/19/83	P,20-C	237.4	280.5	268.7	282.1	301.3	370.6	20.8	133.2
21624	1/19/83	P,20-C	231.9	259.8	255.1	263.9	282.2	333.8	22.4	101.9
21625	1/20/83	P,20-C	257.9	281.0	289.0	300.1	315.2	355.0	34.2	97.1
21626	1/29/83	P,20-C	248.8	288.6	292.6	299.7	329.3	393.8	40.7	145.0
Mean			240.93	271.81	267.73	280.87	301.56	359.67	29.74	118.74
S.D.			10.348	12.006	17.535	14.384	17.119	19.150	10.317	18.776
			Group 4 - 37.5 mg/kg							
21627	1/18/83	P,20-C	274.8	298.3	272.5	273.9	284.6	358.5	-13.7	83.7
21628	1/18/83	P,15-FD	269.3	300.3	275.6	241.2	(219.0)[a]			
21629	1/19/83	P,20-A	240.6	264.8	264.1	244.7	214.1	(161.9)	(-50.7)	(-78.7)
21630	1/19/83	P,16-FD	246.3	274.8	263.5	255.0	225.2[a]		-21.1	
21631	1/19/83	P,17-FD	232.7	259.3	257.5	228.6	203.9	(196.3)[a]	-55.4	
21632	1/19/83	P,20-C	237.8	264.5	257.4	230.5	212.1	275.5	-52.4	37.7
21633	1/29/83	NP,20-C	(281.2)	(299.6)	(280.0)	(246.6)	(215.4)	(251.8)		
Mean			250.25	263.63	265.10	245.65	227.98	317.00	-35.65[S-]	60.70[S-]
S.D.			17.535	21.958	7.558	16.890	32.551	58.690	21.324	32.527

Appendix 1 - Continued
Individual and Mean Maternal Body Weights and Weight Changes
Pilot Rat Teratology Study of T-3352

Maternal Number	Date of Mating	Disposition	Body Weight (grams) - Day of Gestation 0	6	8	12	16	20	Weight Change (grams) Days 6-16	Days 0-20
			Group 5 - 75 mg/kg							
21634	1/18/83	P, 16-FD	251.7	273.4	258.2	226.3	192.7[a]		-80.7	
21635	1/18/83	P, 14-FD	215.6	249.6	233.4	182.6	(171.6)[a]			
21636	1/18/83	P, 13-FD	258.1	251.0	254.0	212.9	(203.4)[a]			
21637	1/19/83	NP, 15-FD	(249.4)	(253.6)	(246.1)	(214.3)	(190.1)[a]			
21638	1/19/83	P, 14-FD	222.7	242.9	237.7	189.3	(171.2)[a]			
21639	1/19/83	P, 15-FD	231.3	266.4	243.1	209.4	(187.6)[a]			
21640	1/21/83	P, 13-FD	254.0	273.5	251.8	220.3	(198.3)[a]			
Mean			238.90	259.47	246.37	206.80				
S.D.			18.020	13.286	9.816	17.331				

Appendix 2
Individual and Mean Daily and Total Food Consumption
Pilot Rat Teratology Study of T-3352

Maternal Number	Food Consumption (gm/day) Day of Gestation 6	8	12	16	20	Total Food Consumption Days 0-20 (grams)[a]
			Group 1 - 0 mg/kg			
21606	16.5	20.0	19.3	20.7	25.0	415.4
21607	20.4	17.4	18.3	19.1	25.4	428.5
21608	19.8	17.5	19.0	18.6	25.1	424.0
21609	15.3	22.6	22.0	19.7	25.2	419.5
21610	18.6	19.2	18.8	20.3	26.2	429.6
21611	19.1	18.6	22.1	21.4	29.9	464.4
Mean	18.28	19.22	19.92	19.97	26.13	430.23
S.D.	1.981	1.933	1.685	1.039	1.895	17.578
			Group 2 - 1 mg/kg			
21613	12.9	19.5	20.7	20.7	26.1	399.2
21614	24.8	24.9	24.3	22.4	31.7	537.3
21615	20.5	20.7	21.7	22.1	27.6	470.1
21616	17.8	16.8	17.9	32.4	28.5	472.7
21617	18.0	18.2	17.8	18.8	19.6	386.7
21618	15.9	15.3	17.9	21.6	26.8	406.7
Mean	18.32	19.23	20.05	23.00	26.72	445.45
S.D.	4.058	3.370	2.665	4.784	3.996	58.101

[a] Statistical group comparisons performed on rank transformed data.

Appendix 2 - Continued
Individual and Mean Daily and Total Food Consumption
Pilot Rat Teratology Study of T-3352

Maternal Number	Food Consumption (gm/day) Day of Gestation 6	8	12	16	20	Total Food Consumption Days 0-20 (grams)[a]
			Group 3 - 10 mg/kg			
21620	16.6	9.0	15.8	12.0	23.4	339.0
21621	17.8	14.1	17.2	15.6	24.2	380.5
21622	19.2	12.1	21.8	20.8	25.6	430.7
21623	18.8	14.5	22.7	19.7	24.3	427.1
21624	17.8	15.3	17.9	17.6	23.0	389.1
21625	14.6	22.9	20.4	20.0	24.7	408.3
21626	15.6	15.7	18.2	18.3	24.6	384.5
Mean	17.20	14.80	19.14	17.71	24.26	394.17
S.D.	1.681	4.246	2.539	3.060	0.860	31.539
			Group 4 - 37.5 mg/kg			
21627	23.2	7.3	9.2	12.1	24.8	361.1
21628	18.8	9.4	2.0	Found dead Day 15		(157.8)
21629	16.6	17.9	55.7	52.7	0.2	586.4
21630	19.5	11.1	9.8	Found dead Day 16		(198.0)
21631	17.3	13.1	7.3	0.3	Found dead Day 17	(177.5)
21632	18.0	14.4	10.4	1.9	20.8	286.8
21633	(17.8)	(6.2)	(2.8)	(0.7)	(13.2)	(203.6)
Mean	18.90	12.20	15.73	16.75	15.27	411.43
S.D.	2.346	3.774	19.816	24.530	13.201	156.013

[a] Statistical group comparisons performed on rank transformed data.
NOTE: Numbers in parentheses are excluded from mean calculation and statistical evaluation.

Appendix 2 - Continued
Individual and Mean Daily and Total Food Consumption
Pilot Rat Teratology Study of T-3352

Maternal Number	Food Consumption (gm/day) Day of Gestation 6	8	12	16	20	Total Food Consumption Days 0-20 (grams)
			Group 5 - 75 mg/kg			
21634	17.5	10.3	10.5	Found dead Day 16		(185.0)
21635	14.7	12.4	1.6	Found dead Day 14		(134.0)
21636	17.4	7.9	1.9	Found dead Day 13		(145.1)
21637	(11.7)	(14.9)	(6.4)	Found dead Day 15, NP		(137.3)
21638	14.1	13.0	1.7	Found dead Day 14		(131.6)
21639	21.8	6.6	31.8	Found dead Day 15		(293.2)
21640	34.7	7.5	3.8	Found dead Day 13		(273.1)
Mean	20.03	9.62	8.55			
S.D.	7.684	2.690	11.886			

NOTE: Numbers in parentheses are excluded from mean calculation and statistical evaluation.

Appendix 3
Individual Body Weights, Gravid Uterine Weights, and
Terminal Body Weights Minus Gravid Uterine Weights
Pilot Rat Teratology Study of T-3352

Maternal Number	Terminal Body Weight (g)	Gravid Uterine Weight (g)	Terminal Body Weight Minus Gravid Uterine Weight (g)
		Group 1 - 0 mg/kg	
21606	367.2	79.3	287.9
21607	366.3	74.1	292.2
21608	374.7	87.5	287.2
21609	376.8	84.9	291.9
21610	358.1	68.7	289.4
21611	356.0	69.3	286.7
Mean	366.52	77.30	289.22
S.D.	8.422	7.925	2.378
		Group 2 - 1 mg/kg	
21613	336.4	36.4	300.0
21614	403.8	90.7	313.1
21615	400.9	90.0	310.9
21616	348.6	68.3	280.3
21617	330.9	73.5	257.4
21618	363.1	51.1	312.0
Mean	363.95	68.33	295.62
S.D.	31.752	21.509	22.459

Appendix 3 - Continued
Individual Body Weights, Gravid Uterine Weights, and
Terminal Body Weights Minus Gravid Uterine Weights
Pilot Rat Teratology Study of T-3352

Maternal Number	Terminal Body Weight (g)	Gravid Uterine Weight (g)	Terminal Body Weight Minus Gravid Uterine Weight (g)
		Group 3 - 10 mg/kg	
21620	344.7	65.7	279.0
21621	359.3	67.7	291.6
21622	360.5	77.9	282.6
21623	370.6	75.5	295.1
21624	333.8	65.7	268.1
21625	355.0	46.8	308.2
21626	399.8	79.1	314.7
Mean	360.53	68.34	291.33
S.D.	20.979	11.081	16.396
		Group 4 - 37.5 mg/kg	
21627	358.5	81.0	277.5
21628	Found dead Day 15		
21629	(161.9) Aborted pups Day 20		
21630	Found dead Day 16		
21631	Found dead Day 17		
21632	275.5	53.8	221.7
21633	(251.8) Not pregnant		
Mean	317.00	67.40	249.60
S.D.	58.690	19.233	39.457

NOTE: Numbers in parentheses are excluded from mean calculation and statistical evaluation.

Appendix 3 - Continued
Individual Body Weights, Gravid Uterine Weights, and
Terminal Body Weights Minus Gravid Uterine Weights
Pilot Rat Teratology Study of T-3352

Maternal Number	Terminal Body Weight (g)	Gravid Uterine Weight (g)	Terminal Body Weight Minus Gravid Uterine Weight (g)
		Group 5 - 75 mg/kg	
21634	Found dead Day 16		
21635	Found dead Day 14		
21636	Found dead Day 13		
21637	Found dead Day 15, Not pregnant		
21638	Found dead Day 14		
21639	Found dead Day 15		
21640	Found dead Day 13		

Appendix 4
Individual and Mean Ovarian, Uterine, and Litter Data
Pilot Rat Teratology Study of T-3352

Maternal Number	Ovarian Corpora Lutea	Uterine Implantations	Implantation Efficiency (%)	Early Resorptions	Resorption Incidences (%)	Number of Fetuses Dead	Number of Fetuses Alive	Fetal Mortality (%)	Fetal Viability (%)
				Group 1 - 0 mg/kg					
21606	14	14	100.0	0	0	0	14	0	100
21607	13	13	100.0	0	0	0	12	0	92.3
21608	(15)[a]	16	a	1	6.3	0	15	0	93.8
21609	17	15	88.2	0	0	0	15	0	100
21610	14	14	100.0	3	21.4	0	11	0	78.6
21611	17	15	88.2	3	20.0	0	12	0	80.0
Mean	15.0	14.5	95.28	1.2	7.95	0	13.2	0	90.78
S.D.	1.87	1.05	6.463	1.47	10.183	0	1.72	0	9.445
				Group 2 - 1 mg/kg					
21613	24	13	54.2	8	61.5	0	5	0	38.5
21614	17	17	100	1	5.9	0	16	0	94.1
21615	15	15	100	0	0	0	15	0	100
21616	13	11	84.6	0	0	0	11	0	100
21617	15	14	93.3	1	7.1	0	13	0	92.9
21618	17	10	58.8	2	20.0	0	8	0	80.0
Mean	16.8	13.3	81.82	2.0	15.75	0	11.3	0	84.25
S.D.	3.82	2.58	20.461	3.03	23.576	0	4.23	0	23.576

[a] Ovary cut - excluded from mean calculation.

Appendix 4 - Continued
Individual and Mean Ovarian, Uterine, and Litter Data
Pilot Rat Teratology Study of T-3352

Maternal Number	Ovarian Corpora Lutea	Uterine Implantations	Implantation Efficiency (%)	Early Resorptions	Resorption Incidences (%)	Number of Fetuses Dead	Number of Fetuses Alive	Fetal Mortality (%)	Fetal Viability (%)
				Group 3 - 10 mg/kg					
21620	14	12	85.7	0	0	0	12	0	100
21621	16	16	100	3	18.8	0	13	0	81.3
21622	15	15	100	1	6.7	0	14	0	93.3
21623	18	15	83.3	1	6.7	0	14	0	93.3
21624	15	14	93.3	3	21.4	0	11	0	78.6
21625	15	9	60.0	0	0	0	9	0	100
21626	16	15	93.8	0	0	0	15	0	100
Mean	15.6	13.7	88.01	1.1	7.66	0	12.6	0	92.36
S.D.	1.27	2.43	13.911	1.35	9.044	0	2.07	0	9.024
				Group 4 - 37.5 mg/kg					
21627	19	18	94.7	2	11.1	0	16	0	88.9
21628	14	13	Found dead Day 15						
21629	14	13	13 Fetuses aborted prior to sacrifice Day 20						
21630	17	17	100	5	Found dead Day 16				
21631	14	13	92.9	2	Found dead Day 17				
21632	19	13	68.4	2	15.4	0	11	0	84.6
21633			Not pregnant						
Mean	16.2	14.5	89.00	2.8	13.25	0	13.5	0	86.75
S.D.	2.48	2.35	14.060	1.50	3.041	0	3.54	0	3.041

Appendix 4 - Continued
Individual and Mean Ovarian, Uterine, and Litter Data
Pilot Rat Teratology Study of T-3352

Maternal Number	Ovarian Corpora Lutea	Uterine Implantations	Implantation Efficiency (%)	Resorptions	Resorption Incidences (%)	Number of Fetuses Dead	Number of Fetuses Alive	Fetal Mortality (%)	Fetal Viability (%)
				Group 5 - 75 mg/kg					
21634	15	11	73.3	Day 16					
21635	a	17	a	Day 14					
21636	16	15	93.8	Day 13					
21637	20		Not pregnant	Found dead Day 15					
21638	11	11	100	Day 14					
21639	17	17	100	Day 15					
21640	15	13	86.7	Day 14					
Mean	15.7	14.0	90.76						
S.D.	2.94	2.76	11.195						

[a] Ovary too autolyzed to count corpora lutea.

Appendix 5
Individual and Mean Live Fetal Data
Pilot Rat Teratology Study of T-3352

Maternal Number	Sex: Number of Males	Sex: Number of Females	Percent Males	Mean Weight (grams): Males Unadjusted	Males Adjusted	Females Unadjusted	Females Adjusted
Group 1 - 0 mg/kg							
21606	7	7	50.0	3.3		3.1	
21607	7	5	58.3	3.6		3.4	
21608	6	9	40.0	3.5		3.2	
21609	9	6	60.0	3.5		3.4	
21610	7	4	63.6	3.4		3.2	
21611	7	5	58.3	3.2		3.2	
Mean	7.2	6.0	55.03	3.42	3.40	3.25	3.25
S.D.	0.98	1.79	8.611	0.147	0.176	0.122	0.157
Group 2 - 1 mg/kg							
21613	2	3	40.0	2.9		3.3	
21614	4	12	25.0	3.3		3.2	
21615	9	6	60.0	3.3		3.2	
21616	9	2	81.8	3.6		3.3	
21617	8	5	61.5	3.4		3.1	
21618	4	4	50.0	3.5		3.3	
Mean	6.0	5.3	53.05	3.33	3.37	3.23	3.22
S.D.	3.03	3.56	19.559	0.242	0.179	0.082	0.158

Appendix 5 - Continued
Individual and Mean Live Fetal Data
Pilot Rat Teratology Study of T-3352

Maternal Number	Sex: Number of Males	Sex: Number of Females	Percent Males	Mean Weight (grams): Males: Unadjusted	Mean Weight (grams): Males: Adjusted	Mean Weight (grams): Females: Unadjusted	Mean Weight (grams): Females: Adjusted
			Group 3 - 10 mg/kg				
21620	7	5	58.3	3.1		3.0	
21621	3	10	23.1	3.0		2.8	
21622	6	8	42.9	3.3		3.2	
21623	9	5	64.3	3.3		2.9	
21624	7	4	63.6	3.6		3.4	
21625	3	6	33.3	3.2		3.1	
21626	11	4	73.7	3.3		3.4	
Mean	6.6	6.0	51.31	3.26	3.27	3.11	3.12
S.D.	2.94	2.24	18.533	0.190	0.176	0.234	0.157
			Group 4 - 37.5 mg/kg				
21627	9	7	56.3	2.9		2.7	
21628	Found Dead Day 15						
21629	Pups absorted prior to sacrifice Day 20						
21630	Found Dead Day 16						
21631	Found Dead Day 17						
21632	6	5	54.5	2.6		2.5	
21633	Not pregnant						
Mean	7.5	6.0	55.40	2.75	2.72S-	2.60	2.60S-
S.D.	2.12	1.41	1.273	0.212	0.176	0.141	0.157

Appendix 5 - Continued
Individual and Mean Live Fetal Data
Pilot Rat Teratology Study of T-3352

Maternal Number	Sex: Number of Males	Sex: Number of Females	Percent Males	Mean Weight (grams): Males: Unadjusted	Mean Weight (grams): Males: Adjusted	Mean Weight (grams): Females: Unadjusted	Mean Weight (grams): Females: Adjusted
Group 5 - 75 mg/kg							
21634	Found Dead Day 16						
21635	Found Dead Day 14						
21636	Found Dead Day 13						
21637	Found Day 15, Not pregnant						
21638	Found Dead Day 14						
21639	Found Dead Day 15						
21640	Found Dead Day 14						

Appendix 6
Individual Visceral Findings for Each Litter
Pilot Rat Teratology Study of T-3352

	Group 1 - 0 mg/kg						Group 2 - 1 mg/kg					
Maternal Number:	2 1 6 0 6	2 1 6 0 7	2 1 6 0 8	2 1 6 0 9	2 1 6 1 0	2 1 6 1 1	2 1 6 1 3	2 1 6 1 4	2 1 6 1 5	2 1 6 1 6	2 1 6 1 7	2 1 6 1 8
Number of fetuses examined by Wilson's technique[a]	3	3	3	3	3	3	3	3	3	3	3	3
Number of fetuses within normal limits	3	3	2	3	3	3	3	2	3	3	3	3
Number of fetuses having one or more visceral variants	0	0	1	0	0	0	0	1	0	0	0	0
Percent of fetuses having one or more visceral variants (%)	0.0	0.0	33.33	0.0	0.0	0.0	0.0	33.33	0.0	0.0	0.0	0.0
Number of fetuses with:												
Dilated renal pelvis								1				
Dilated ureter(s)			1									
Number of fetuses having one or more visceral anomalies	0	0	0	0	0	0	0	0	0	0	0	0
Percent of fetuses having one or more visceral anomalies (%)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

	Group 3 - 10 mg/kg							Group 4 - 37.5 mg/kg	
Maternal Number:	2 1 6 2 0	2 1 6 2 1	2 1 6 2 2	2 1 6 2 3	2 1 6 2 4	2 1 6 2 5	2 1 6 2 6	2 1 6 2 7	2 1 6 3 2
Number of fetuses examined by Wilson's technique[a]	3	3	3	3	3	3	3	3	3
Number of fetuses within normal limits	3	3	3	3	3	3	3	2	1
Number of fetuses having one or more visceral variants	0	0	0	0	0	0	0	1	2
Percent of fetuses having one or more visceral variants (%)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	33.33	66.67
Number of fetuses with:									
Dilated renal pelvis									1
Dilated ureter(s)								1	1
Incompletely descended testis(es)									
Number of fetuses having one or more visceral anomalies	0	0	0	0	0	0	0	1	1
Percent of fetuses having one or more visceral anomalies (%)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	33.33	33.33
Number of fetuses with:									
Cleft palate								1	1

[a] Three fetuses from each litter.

Appendix 7
Individual Skeletal Findings for Each Litter
Pilot Rat Teratology Study of T-3352

	Group 1 - 0 mg/kg						Group 2 - 1 mg/kg					
Maternal Number:	2 1 6 0 6	2 1 6 0 7	2 1 6 0 8	2 1 6 0 9	2 1 6 1 0	2 1 6 1 1	2 1 6 1 3	2 1 6 1 4	2 1 6 1 5	2 1 6 1 6	2 1 6 1 7	2 1 6 1 8
Number of fetuses examined	3	3	4	4	4	4	2[a]	3	3	3	3	3
Number of fetuses within normal limits	0	2	1	1	1	0	2	1	0	2	0	1
Number of fetuses having one or more skeletal variants	3	1	2	2	2	3	0	2	3	1	3	2
Percent of fetuses having one or more skeletal variants (%)	100.00	33.33	66.67	66.67	66.67	100.00	0.0	66.67	100.00	33.33	100.00	66.67
Number of fetuses with:												
Lagging Ossification												
Skull - incomplete closure (less than 75%)				2	2				2			
- interparietal ossification incomplete	2			2	2	2			1			1
- supraoccipital ossification incomplete	3		1	2	2	1		2	2		3	
- hyoid ossification incomplete				1								
- hyoid nonossified	2	1	2	1	2	1			2			
- parietals irregularly ossified	1			1	2				3			
Rib cage - sternebrae bipartite												1
- ribs irregularly ossified					1				1			
Vertebral column - thoracic centra ossification incomplete		1										
- thoracic centra nonfused	1									1		
- caudal less than three ossified						1						
Bone Alignment												
Extra rib(s)				1								
Angulated ribs				1	1				1			
Number of fetuses having one or more skeletal anomalies	0	0	0	0	0	0	0	0	0	0	0	0
Percent of fetuses having one or more skeletal anomalies (%)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

[a] One fetus was damaged during preparation so number of metatarsals and phalanges could not be determined.

Appendix 7 - Continued
Individual Skeletal Findings for Each Litter
Pilot Rat Teratology Study of T-3352

	Group 3 - 10 mg/kg							Group 4 - 37.5 mg/kg	
	2	2	2	2	2	2	2	2	2
	1	1	1	1	1	1	1	1	1
	6	6	6	6	6	6	6	6	6
Maternal	2	2	2	2	2	2	2	2	3
Number:	0	1	2	3	4	5	6	7	2
Number of fetuses examined	3	3	3	3	3	3	3	3	3
Number of fetuses within normal limits	2	0	2	2	2	2	0	1	0
Number of fetuses having one or more skeletal variants	1	3	1	1	1	1	3	2	3
Percent of fetuses having one or more skeletal variants (%)	33.33	100.00	33.33	33.33	33.33	33.33	100.00	66.67	100.00
Number of fetuses with:									
Lagging Ossification									
Skull - incomplete closure (less than 75%)				1					1
- interparietal ossification incomplete		3		1			2	1	1
- supraoccipital ossification incomplete		3		1			2	1	2
- hyoid nonossified		1		1			1		
- parietals irregularly ossified		1		1	1		2		
Rib cage - sternebrae bipartite	1						1	1	3
Vertebral column - thoracic centra ossification incomplete			1						
- thoracic centra nonfused						1			
- caudal less than three ossified						1			1
Bone Alignment									
Extra rib(s)				1					
Angulated ribs									
Number of fetuses having one or more skeletal anomalies	0	0	0	0	0	0	0	0	0
Percent of fetuses having one or more skeletal anomalies (%)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

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Appendix 8
References
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STATISTICAL METHODS/LABORATORY PROCEDURES

Bartlett, M. S., "Some Examples of Statistical Methods of Research in Agriculture and Applied Biology." J. Royal Statist. Soc. Suppl., IV:137-183, 1937.

Draper, N. R. and W. G. Hunter (1969), "Transformations: Some Examples Revisited", Technometrics, 11:23-40.

Dunnett, C. W., "A Multiple Comparison Procedure for Comparing Several Treatments with a Control." J. Am. Stat. Assoc., 50:1096-1121, 1955.

Dunnett, C. W., "New Tables for Multiple Comparisons with a Control." Biometrics, 20:482-491, 1964.

Games, P. A., and Howell, J. F., "Pairwise Multiple Comparison Procedures with Unequal N's and/or Variances: A Monte Carlo Study." J. Ed. Statist., 1:113-125, 1976.

Levene, H., "Robust Tests for Equality of Variances," in I. Olkin edited, Contributions to Probability and Statistics, Stanford University Press, Palo Alto, 1960.

Staples, R. E. and Schnell, V. L., "Refinements in Rapid Clearing Technic in the KOH-Alizarin Red S Method for Fetal Bone." Stain Technology, 39:61-63, 1964.

Wilson, J. G. and Warkany, J., editors. Teratology: Principles and Techniques, The Univ. of Chicago Press, Chicago, 1965, pp. 251-277.

Winer, B. J., Statistical Principles in Experimental Design, 2nd Ed., McGraw-Hill, N.Y., Chs. 3-10, 1971.