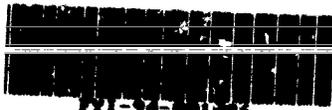


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EXXON CHEMICAL AMERICAS



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Safety and Environmental Affairs Department
Safety Programs
L. E. Schmeltz
MANAGER

FYI-0295-001086

February 2, 1995

Re: TSCA Section 8(e) FYI Notice FYI-OTS-0392-0835

U. S. Environmental Protection Agency
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Dear Sir or Madam:

On October 8, 1993 Exxon Chemical Americas submitted a final copy of a report "INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE WITH DIMETHYLCARBONATE" as additional information for the earlier March 11, 1992 Section 8(e) FYI submission.

We have been advised by Mr. Paul McMann of your office that as a result of an FOI request which was filed to request a copy of the report, EPA cannot locate that information. Mr. McNiann requested that Exxon Chemical Americas provide another copy for the EPA files.

Enclosed is another copy of our 419 page report. If there are questions, I can be reached at (713) 870-6874.

Very truly yours,

Harry L. Hunter, Jr.

Harry L. Hunter, Jr.

Attachment

cc: W/O Attachment
D. D. Sigman

P.O. Box 3272, Houston, Texas 77253-3272
Tel: (713) 870-6212 Fax: (713) 870-6661

A Division of Exxon Chemical Company, A Division of Exxon Corporation



Sample No. 91-073

EXXON BIOMEDICAL SCIENCES, INC.

FINAL REPORT

PROJECT NUMBER: 107334

TEST MATERIAL: MRD-91-073

**INHALATION DEVELOPMENTAL TOXICITY STUDY
IN MICE WITH DIMETHYLCARBONATE**

PERFORMED FOR:

**EXXON CHEMICAL COMPANY
1900 East Linden Avenue
Linden, New Jersey 07036**

PERFORMED AT:

**EXXON BIOMEDICAL SCIENCES, INC.
Toxicology Laboratory
Mettlers Road
CN 2350
East Millstone, New Jersey 08875-2350**

COMPLETION DATE: July 28, 1992

0004

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**INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DINITHYLCARBONATE (MRD-91-073): 107334**

APPROVAL SIGNATURES

Fred T. Whitman 28 JUL 92

F. T. Whitman, B.A. Date
Inhalation Supervisor

Richard D. Phillips 28-Jul-92

R. D. Phillips, Ph.D. Date
Mammalian Toxicology Laboratory Director

CERTIFICATION OF GOOD LABORATORY PRACTICES

I hereby declare that, to the best of my knowledge, this study was conducted in compliance with U.S. EPA Good Laboratory Practice Regulations (40 CFR, Part 792), U.S. EPA TSCA test guidelines for inhalation developmental toxicity studies (40 CFR, Part 798), and the OECD Guideline for Testing of Chemicals, "Teratogenicity" (Section 414, adopted May 12, 1981).

Bruce K. Beyer 28-Jul-92

B. K. Beyer, Ph.D. Date
Study Director

**INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

QUALITY ASSURANCE STATEMENT

STUDY NUMBER: 107334

TEST SUBSTANCE/ARTICLE: MRD-91-073

STUDY SPONSOR: EXXON CHEMICAL COMPANY

Listed below are the dates that this study was inspected by the Quality Assurance Unit of Exxon Biomedical Sciences, Inc., and the dates findings were reported to the Study Director and Management.

<u>Date(s) of Inspection</u>	<u>Reported to Study Director</u>	<u>Reported to Management</u>
14-Nov-91	25-Nov-91	02-Dec-91
22-Nov-91	26-Nov-91	02-Dec-91
09-Dec-91 03,14-Jan-92	21-Jan-92	03-Feb-92 04-Feb-92
18-29-May-92 12-17-Jun-92	21-Jul-92	28-Jul-92


Joanne R. Jackson, B.S.
Quality Assurance Analyst


Date

PERSONNEL

Study Director:	B. K. Beyer, Ph.D.
Sponsor Representative:	C. J. Bevan, Ph.D.
Inhalation, Compound Preparation and Necropsy Supervisor:	F. T. Whitman, B.A.
Inhalation Research Technician:	J. J. Signorin, B.S.
Report Preparation Supervisor:	E. R. Frank, B.A.
Toxicology and Animal Care Supervisor:	R. C. Forgash, B.S.
Veterinarian:	R. L. Harris, D.V.M.
Quality Assurance Supervisor:	J. R. Jackson, B.S.
Maintenance Supervisor:	J. L. McGrath, A.S.
Consultant:	S. B. Harris, Ph.D.
Statistician:	M. J. Nicolich, Ph.D.

**PERINATAL DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

SUMMARY

The objective of this study was to determine the potential of dimethylcarbonate (MRD-91-073) to induce fetal structural and/or other abnormalities when administered to the mouse during pregnancy. The test material was administered in the breathing air to three groups of 96 female mice each at doses of 300, 1000, and 3000 ppm. An additional group of 96 female mice served as a chamber control group. EBSI Study T00034M2 was performed concurrently with EBSI Study 107334 and provided an untreated group of 96 female mice as a control for stress-induced abnormalities. Animals allocated to Study T00034M2 remained in the study room with unlimited access to food and water during exposures for the treated groups. The exposed animals were placed into appropriate inhalation chambers which were operated under dynamic conditions. The exposure period was 6 hours per day plus time for equilibration (theoretical T99). Mated females were exposed daily from GD 6 (GD = gestation day) through GD 15.

Males and females were paired based on sequential Permanent Identification Number (PIN) and housed overnight until confirmation of mating by observation of a copulatory plug in the vagina or cage pan (plug = GD 0). Each mated female then was returned to its own cage and new females were placed in the males' cages until the required number of mated females was obtained. Mated females were assigned to dose groups in the order of mating.

Clinical observations were made prior to selection and daily thereafter. The animals were examined for viability at least twice daily during the treatment period and at least once daily at other times during the study. In-chamber observations for mortality and obvious toxic signs were made on the animal population as a whole during approximately the first and last hours of daily exposures. Body weights were taken prior to selection, and on GD 0, 6, 9, 12, 15, and 18. Food consumption was measured concurrently with body weights during gestation.

On GD 18, maternal animals were euthanized and cesarean sections were performed on the first 30 pregnant animals per group. Any additional animals per group were discarded without further examination of the dams or their litters. A cursory gross necropsy was performed on maternal animals and uterine plus ovarian weight was measured. Body weight was recorded on the day of necropsy. Uterine contents were examined and required uterine data collected. Live fetuses were euthanized by placing a drop of sodium pentobarbital on the tongue. Each live fetus was weighed and examined externally for gross malformations, including cleft palate. The sex of each fetus was determined by external examination

**INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107224**

and confirmed internally on those fetuses receiving visceral examinations. Approximately one-half of the fetuses of each litter were decapitated and the heads were examined for the presence of abnormalities. The viscera of all fetuses to be decapitated also were examined for variations and malformations. The remaining fetuses were eviscerated and processed for skeletal staining and examined for the presence of malformations and ossification variations.

There were no remarkable differences between untreated control and chamber control maternal or fetal data collected during the study. There were no treatment-related clinical in-life observations, in-chamber toxic signs, or mortality in any dose group. One high dose dam was euthanized on GD 12 for humane reasons following a mechanical injury.

Statistically significant maternal body weight gain suppression and decreased food consumption occurred in the high dose group when compared with chamber controls during the majority of body weight and food consumption intervals, including the treatment period (GD 6-15) and overall gestation period (GD 0-18).

The majority of dams had no observable abnormalities at postmortem examination. Incidental findings observed at necropsy were limited to the untreated control and chamber control groups. Statistically significant decreases in mean live and mean male fetuses and a statistically significant increase in mean resorptions were observed in the high dose group when compared with chamber controls.

Mean fetal body weights in males and females in the high dose group were statistically significantly reduced when compared with those of chamber controls. Fetal variations and malformations were observed in all groups, including untreated and chamber controls. However, the incidences of total external, visceral, and skeletal malformations were statistically significantly higher in the high dose group when compared with chamber controls on both a per fetus and per litter basis. A treatment-related increase in the number of rib ossification sites was observed in the high dose group compared with chamber controls and was associated with increased incidences of rudimentary and supernumerary ribs.

In conclusion, maternal toxicity was observed at a dose level of 3000 ppm, as indicated by maternal body weight gain suppression and reduced food consumption compared with controls. Accordingly, the maternal NOAEL (No Observable Adverse Effect Level) was established at 1000 ppm of dimethylcarbonate (MRD-91-073) under the conditions of this study. Fetal body weights were reduced in the high dose group compared with controls. In addition, there was an increased incidence of fetal malformations and developmental variations

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334

in the high dose group compared with chamber controls. Therefore, the fetal NOEL was established at 1000 ppm of dimethylcarbonate under the conditions of this study. Teratogenic effects characterized by cleft palate, microtia, low set ears, multiple skull bone malformations, and fused vertebral arches were observed at a maternally-toxic exposure level of 3000 ppm when administered under the conditions of this study.

OBJECTIVE

This study was conducted to determine the potential of the test material, dimethylcarbonate, to induce fetal structural and/or other abnormalities when administered to the mouse by inhalation during pregnancy.

This study was conducted for Exxon Chemical Company, 1900 East Linden Avenue, Linden, N.J. 07036 (subsequently referred to as the Sponsor) on the test material, designated MRD-91-073, in the mouse.

This study was performed at Exxon Biomedical Sciences, Inc. Toxicology Laboratory (an AAALAC-accredited facility), Mettlers Road, CN 2350, East Millstone, New Jersey 08875-2350.

Study Initiation

November 13, 1991

Biophase Test Period

November 13, 1991 to December 10, 1991

Experimental Termination

February 3, 1992

MATERIALS AND METHODS

TEST MATERIAL

Material Identification

Sponsor Identification: Dimethylcarbonate
Exxon Biomedical Sciences Identification: MRD-91-073
Batch Number: III
Date Received: October 21, 1991
Description: Colorless liquid
Assumed to be 100% pure for purposes of dosing.
Storage Condition: Room temperature

**INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

Characterization of Test Material

The methods of synthesis, fabrication, and/or derivation of the test material are documented by the Sponsor and are maintained at Exxon Chemical Company, 1900 East Linden Avenue, Linden, N J. 07036.

Analysis of Test Material

The stability, identity, strength, and composition or other characteristics which will appropriately identify the test material are documented by the Sponsor and are maintained at Exxon Chemical Company, 1900 East Linden Avenue, Linden, N.J. 07036.

It is unknown whether these analyses were determined and documented in a GLP compliant manner. This is a deviation from Good Laboratory Practices (U.S. EPA TSCA).

Analysis of Mixture

Not applicable

Sample Retention

Two archival samples of undiluted MRD-91-073 were collected by the Compound Preparation Department and stored at room temperature.

Carrier

Air.

TEST SYSTEM

Test Animal

Species: Mouse
Strain/stock: CD-1
Supplier (Females): Charles River Breeding Labs, Inc.
Portage, Michigan

Animal Receipt Information

Females:

Receipt Date: October 28, 1991
Purchase Order Number: 1GM38201

Quarantine and Acclimation Period

Females: 17 Days; Examined once daily.

Number and Sex

384 sexually mature virgin females
120 sexually mature males (maximum)

Age at Initiation of Mating

Females: Approximately 68 days

Weight at Designated Day 0 of Gestation

Females: 24.85 to 33.34 grams

Animal Identification

Monel ear tags and corresponding cage identification

Justification for Selection of Test System

The mouse is among the species of choice for developmental toxicity testing according to U.S. EPA TSCA test guidelines for inhalation developmental toxicity studies (40 CFR Part 798) and the OECD Guideline for Testing of Chemicals, "Teratogenicity" (Section 414, adopted May 12, 1991).

Selection

More animals than required for the conduct of the study were purchased and acclimated. Animals determined to be unsuitable for inclusion on this study because of poor health, outlying body weights, or other abnormalities were excluded from selection by the Staff Veterinarian, Study Director, and/or technical staff.

Mating

After approximately 3:00 p.m., on the initial scheduled mating day, females were placed in males' cages in a 1:1 (male:female) ratio. Males and females were paired based on sequential PIN (Permanent Identification Number). A suitable number of animals were co-housed in an attempt to produce a suitable number of mated females. Mating was confirmed on the following morning by observation of a copulatory plug (vaginal or in the cage pan). The day on which mating was confirmed was the female's Day 0 of gestation (GD 0). After confirmation of mating, each mated female was returned to its own cage. New females were placed in the males' cages until the required number of mated females was obtained by continuous cohabitation in consideration of laboratory scheduling. Mated females were assigned to dose groups in the order of mating. Accordingly, the first confirmed mated female was assigned to Group 1, the next to Group 2, and so on until all mated animals for a given day had been assigned to dose groups. On subsequent days, the next group in sequence was filled by the first confirmed mated on that day, and so on. Mated females which were assigned to Study T00034M2 (untreated controls) were assigned as if they were in Group 5 of Study 107734 for purposes of group allocation only. Assignments were made until all confirmed mated females were assigned to groups, or until as many were assigned as could be examined on GD 18. Upon completion of the mating period, all males and unassigned females either were euthanized and discarded or transferred to the stock colony maintained by the Developmental Toxicology Group.

Husbandry

Housing

Room numbers: 522 (October 28 - November 13, 1991),
505 (November 13 - December 10, 1991)
Housing: Single housed during the test period
Caging: Suspended stainless steel and wire mesh
with absorbent paper below cages.

Feed

Purina Certified Rodent Chow (5002 Meal), ad libitum.
Manufacturer: Purina Mills, Inc.
Richmond, Indiana

Analysis: As provided by Purina. Copies of the
feed analyses are maintained in the EBSI
Toxicology Laboratory Archives.

Contaminants: There were no known contaminants in the
feed believed to have been present at
levels that may have interfered with this
study.

Water

Automatic watering system, ad libitum

Supplier: Elizabethtown Water Company
Elizabeth, New Jersey

Analysis: Periodic analysis is provided by Elizabethtown
Water Company. Copies of these analyses are
maintained in the EBSI Toxicology Laboratory
Archives.

Contaminants: There were no known contaminants in the
water believed to have been present at
levels that may have interfered with
this study.

NOTE: Animals were without food or water while in the
exposure chambers.

**INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

Environmental Conditions

Temperature: 68 to 76°F

Humidity: 40 to 70% relative humidity (RH)

Lighting: Approximately 12 hours light and 12 hours dark by automatic timer. Diurnal cycle approximately 0700 to 1900 hours; nocturnal cycle approximately 1900 to 0700 hours.

Monitored at least once daily.

Chamber Temperature Range: 68 to 76°F

Chamber Humidity Range: 40 to 60% RH, if practical due to the physical characteristics of the test material and generation method.

EXPERIMENTAL DESIGN

Preparation of Test Material

The test material was administered as received from the Sponsor.

Experimental Groups

Study	Group	Description	Number of Mated Females/Group*	Target Exposure Levels (ppm)
T00034M2	1	Untreated control	96	0
107334	1	Chamber control	96	0
	2	Low	96	300
	3	Intermediate	96	1000
	4	High	96	3000

*Maximum number. NOTE: Only the first 30 pregnant animals per group were examined at scheduled cesarean section on GD 18. Once this number of pregnant animals had been reached, the in-life portion of the particular group was terminated and all animals which remained on study in that group at that point were euthanized and discarded without further examination of dams or their litters. Similarly, mating and/or inhalation exposures were terminated at that point.

Administration of Test Material and Exposure Schedule

The experimental and chamber control animals were placed into appropriate inhalation chambers which were operated under dynamic conditions. The exposure period was 6 hours per day plus time for equilibration (theoretical T99). Mated females were exposed daily from GD 6 through GD 15.

Justification of Dosing Route

Inhalation is an accepted route of administration according to U.S. EPA TSCA and OECD regulations and represents the most likely route of human exposure.

**INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRL-91-073): 107334**

The Chamber

The chambers used for exposure were stainless steel and glass or plastic and had a total volume of approximately 1000 liters. They were operated at a flow rate sufficient to ensure timely equilibration (approximately 12-15 air changes/hour). The flow of air through each chamber was monitored continuously using a calibrated flow meter and recorded approximately every 30 minutes. All chambers were maintained at a slight negative pressure.

The Test Atmosphere

The test material was administered in the breathing air of the animals. The method of test atmosphere generation is described in Appendix I.

Analytical Procedures

Nominal Concentration

A nominal exposure concentration was calculated. The weight of the test material used was divided by the total volume of air passing through the chamber to give the nominal concentration.

Actual Concentration

Determinations of actual chamber concentration were made approximately once each hour by an appropriate, validated analytical technique. The method and results are described in Appendix I.

Chamber Homogeneity

During the trials for the rangefinding study (EBSI Study 107333), samples were drawn from at least four points in the chamber to ascertain the homogeneity of the test material distribution. The method and results are described in Appendix I.

Experimental Evaluation

The animals were examined for viability at least twice daily during the treatment period and at least once daily at other times during the study. Animals appearing moribund were euthanized at the discretion of the Study Director or his designee.

Body weights were recorded prior to selection and for each mated female assigned to a study group, on GD 0, 6, 9, 12, 15, and 18.

Food consumption was measured concurrently with body weights during gestation.

A clinical examination was given to each female prior to selection and daily thereafter.

In-chamber observations for mortality and obvious toxic signs were made on the animal population as a whole during approximately the first and last hours of daily exposures.

Euthanasia and Cesarean Section

Moribund animals were euthanized at the discretion of the Study Director or his designee. Surviving mated animals were euthanized on GD 18 by carbon dioxide asphyxiation and exsanguination. All females assigned to groups were examined by a cursory gross necropsy.

Uterine weight with ovaries attached was recorded at the time of necropsy. Body weights were recorded on the day of necropsy. Uterine contents were examined and the numbers and locations of implantation sites, early and late resorptions, live, dead, and externally malformed fetuses were recorded. The uteri of all apparently non-pregnant dams were stained to confirm pregnancy status.

Examination of Fetuses

Each live fetus was weighed and examined externally for gross malformations, including cleft palate. All live fetuses were euthanized by placing a drop of sodium pentobarbital on the tongue prior to internal examination. The sex of each live fetus was determined by external examination and confirmed internally only on those fetuses receiving visceral examinations.

Approximately one-half of the fetuses of each litter were decapitated after being euthanized. These heads were preserved in Bouin's solution for at least two weeks, then rinsed and subsequently stored in 70% ethanol. Free-hand razor blade sections of the Bouin's-fixed fetal heads were examined for the presence of abnormalities. The viscera of all fetuses to be decapitated also were examined by fresh dissection. The remaining live fetuses were eviscerated after being euthanized, processed for skeletal staining with Alizarin red, and examined for the presence of malformations and ossification variations.

**INEHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

Representative malformations were photographed at the discretion of the Study Director.

Records

The study protocol, final report, specimens, photographs, and all raw data are maintained on file in the EBSI Toxicology Laboratory Archives. All computer programs used for data collection and report generation also are stored in the EBSI Toxicology Laboratory Archives.

ANALYSIS OF DATA AND STATISTICAL METHODS

The following parameters were analyzed statistically for significant differences from corresponding chamber control mean values:

- mean body weight and gravid uterine weight of pregnant animals
- mean body weight change of pregnant animals
- mean food consumption of pregnant animals
- mean number of implantations per dam
- mean number of resorptions per dam
- mean number of live fetuses per litter
- mean number of live male fetuses per litter
- mean number of live female fetuses per litter
- mean number of dead fetuses per litter
- mean number of malformed fetuses per litter
- mean number of affected (resorbed, dead or malformed) offspring per litter
- mean number of fetuses with developmental variations per litter
- mean post-implantation loss

Live fetuses as fraction of implants, dead fetuses as fraction of implants, and resorptions as fraction of implants were calculated for each litter. Fractions were transformed by Cochran's transformation followed by the arc sine transformation. The raw fractions and the transformed fractions were tested for statistical significance.

The following statistical methods were employed:

Bartlett's test of homogeneity of variance was used to determine if the groups had equivalent variances at the 1% level of significance (Snedecor and Cochran, 1971).

1. If the variances were equivalent, the hypothesis that there was no difference in response between the groups was tested using a standard one-way analysis of variance (Snedecor and Cochran, 1971). If the ANOVA was significant, Dunnett's test was performed to determine which treated groups differed from the chamber control group (Dunnett, 1964). A linear regression to test for a dose response also was performed and tested for lack of fit to the regression (Snedecor and Cochran, 1971).

2. If the variances were not equivalent, then a Kruskal-Wallis (non-parametric) test was performed to determine if the treatment effects were equivalent (Hollander and Wolfe, 1973). If there was a difference, Dunn's Summed Rank Test was used to determine which treatment groups differed from the chamber control group (Hollander and Wolfe,

**REPRODUCTION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

1973). Jonckheere's test for ordered response also was performed (Hollander and Wolfe, 1973).

The Student t-test was used to compare gestation body weights, food consumption, and uterine implantation indices between the untreated and chamber control groups in order to evaluate the effects of stress placed on the animals while in the exposure chambers (Dixon and Massey, 1969).

Fetal weight was analyzed by a standard nested analysis of covariance with fetuses nested within dams and with dams nested within doses, and litter size (both sexes combined) as the covariate (Snedecor and Cochran, 1971). If differences in groups were identified, the Least Significant Difference (LSD) technique was used to determine which groups differed from the chamber control group (Snedecor and Cochran, 1971). Male and female pups were tested separately (the covariate was combined sexes in each analysis). All tests were reported at the 5% or 1% level of significance.

The following were calculated and analyzed for statistical significance as described below:

- incidences of individual and total malformations
- incidences of individual and total variations

A standard chi-square analysis was performed to determine if the proportion of incidences differed between the groups tested (Snedecor and Cochran, 1971). Each treatment group was then compared to the chamber control group using a 2 x 2 Fisher Exact test (Bradley, 1968). Armitage's test for linear trend in the dosage groups was performed (Snedecor and Cochran, 1971).

RESULTS

A. MATERNAL OBSERVATIONS

1. CLINICAL IN-LIFE OBSERVATIONS

A summary of clinical in-life observations is presented in Table 1. The majority of dams were free of any observable abnormalities during gestation. Incidental findings which were considered unrelated to treatment included single or low incidences of eye damage, necrotic tail, or red vaginal material in the untreated control group, alopecia in the chamber control group, and red, green, or yellow vaginal material and anogenital staining in the high dose group. One high dose dam had a broken leg and was euthanized on GD 12.

One high dose dam delivered two live and eight dead offspring on presumed GD 14. Since the offspring appeared to correspond in size and development to those normally observed on GD 18, the mating confirmation for this dam was considered incorrect. This dam subsequently was euthanized. One dam each in the untreated control and low dose groups and two dams in the chamber control group delivered offspring on GD 18 prior to scheduled cesarean sections. Four untreated control females were not pregnant. Two females each in the chamber control and mid-dose groups and three females each in the low and high dose groups were not pregnant. Individual gestation observations are presented in Appendix A.

In-chamber observations:

Mortality and obvious toxic signs were not observed in the animal population as a whole during the first or last hours of daily exposures.

2. GESTATION BODY WEIGHTS

Mean gestation body weights and body weight change are presented in Table 2. There were no meaningful differences observed in mean gestation body weight or uterine weight between the untreated control and chamber control groups at any body weight interval or after correction for gravid uterine weight (GD 18C). There were no differences in body weight change between the untreated and chamber control groups during either the treatment interval (GD 6-15) or the overall gestation period (GD 0-18), even after correction for gravid uterine weight (GD 0-18C).

Mean gestation body weights and uterine weights were similar between chamber control and treated groups at doses of 1000

**REPRODUCTION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334**

ppm and lower for all body weight intervals and after correction for gravid uterine weight. There were statistically significant decreases in high dose mean body weight (GD 15, GD 18 and GD 18C) and mean uterine weight compared with chamber controls. Statistically significant body weight gain suppression was observed in the high dose group at all intervals from GD 9 through GD 18, during the treatment period (GD 6-15), and the overall gestation period (GD 0-18), and after correction for gravid uterine weight (GD 0-18C). Individual body weight data are presented in Appendix B.

3. GESTATION FOOD CONSUMPTION

Mean gestation food consumption data are presented in Table 3. There were no differences observed between untreated control and chamber control dams at any food consumption interval during gestation, including the treatment interval (GD 6-15) and overall gestation period (GD 0-18).

Statistically significant decreases in mean gestation food consumption were observed in the low dose group when compared with controls during the GD 6-9 interval and in the mid-dose group when compared with chamber controls during the GD 9-12 interval, the treatment period and overall gestation period. Statistically significant decreases also were observed in the high dose group when compared with controls at all food consumption intervals after GD 6, including during the treatment period and overall gestation period. Individual food consumption data are presented in Appendix C.

4. MATERNAL NECROPSY OBSERVATIONS

A summary of maternal necropsy observations is presented in Table 4. All dams in treated groups which survived to scheduled cesarean section had no remarkable postmortem abnormalities. Incidental postmortem findings were limited to untreated and chamber control dams and included single or low incidence of eye damage, necrotic tail, and fluid-filled sacs surrounding the ovaries. The high dose dam that was euthanized on GD 12 was observed with an extremely swollen left hindleg. Individual necropsy observations are presented in Appendix D.

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5. UTERINE IMPLANTATION DATA

Mean uterine implantation data are presented in Table 5. There were no meaningful differences between untreated control and chamber control dams for any uterine implantation indices. There was a decrease in post-implantation loss observed in the chamber control group when compared with the untreated control group. Since all other uterine implantation parameters were similar between untreated control dams and chamber control dams, this finding was considered incidental.

Treatment-related effects on uterine implantation indices were observed in the high dose group when compared with the chamber control group. In the high dose group, statistically significant decreases were observed in mean number of live fetuses per litter and mean number of male fetuses per litter, and a statistically significant increase was observed in mean number of resorptions per litter when compared with the chamber control group. Accordingly, mean number of fetuses/implantations was decreased and mean number of resorptions/implantations was increased in the high dose group when compared with the chamber control group. In addition, statistically significant increases in mean post-implantation loss, mean number of malformed fetuses per litter, and mean number of affected fetuses per litter were observed in the high dose group when compared with the chamber control group. There were no statistically significant differences in uterine implantation indices observed between the chamber control group and treated groups exposed to 1000 ppm or below. Individual uterine implantation data are presented in Appendix E.

B. FETAL OBSERVATIONS

1. FETAL BODY WEIGHTS

Mean fetal body weights are presented in Table 6. There were no differences in body weights between untreated control and chamber control male or female fetuses. Statistically significantly reduced body weights were observed in high dose male and female fetuses when compared with corresponding chamber control fetuses. Individual fetal body weights are presented in Appendix F.

2. FETAL VARIATIONS AND MALFORMATIONS

Summaries of developmental variations and fetal malformations are presented in Tables 7 and 8, respectively. There were no remarkable differences in total developmental variations or malformations observed between untreated control and chamber

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control groups on either a per fetus or per litter basis. There was a statistically significantly increased incidence of an extra site of ossification between the frontal bones of chamber control mice compared with untreated controls. The biological significance of this finding is unknown but presumably was related to generalized stress associated with inhalation exposures. A statistically significant reduction was observed in the incidence of fetuses with well-formed lumbar ribs in the chamber control group compared with the untreated control group. However, the litter incidences of this developmental variation were similar between the two control groups. In the absence of effects on other fetal parameters in these groups, this finding was considered incidental.

There were no biologically meaningful differences in total external or visceral variations in treated groups compared with chamber controls, although statistically significant reductions in total external variations were observed in the low and mid-dose groups when compared with controls on a per fetus and per litter basis. A statistically significant increase in the total number of fetuses with skeletal variations was observed in the high dose group when compared with chamber controls and there was a statistically significantly increased incidence of stunted fetuses in the high dose group when compared with chamber controls on both a per fetus and per litter basis. External variations included statistically significantly decreased incidences of abnormal flexure of the hindlimb(s) in the low, mid-, and high dose groups when compared with chamber controls on both a per fetus and per litter basis. Fleshy tab(s) at the side(s) of the mouth were observed in six fetuses of one litter in the high dose group and was statistically significant only on a per fetus basis when compared with the chamber control group. In addition, protruding tongue was observed in association with anencephaly or exencephaly in two high dose fetuses.

Skeletal variations were observed throughout the groups and consisted primarily of hypoplastic or misshapen elements of the skull, sternbrae, ribs, vertebrae or pelvic girdle. Supernumerary bones or sites of ossification were observed in elements of the skull, sternbrae, vertebrae and hindpaw. Statistically significant increases were observed on a per fetus basis in misshapen squamosal or pubis in the high dose group when compared with chamber controls and a statistically significant decrease was observed on a per fetus basis in asymmetric sternbrae in the low dose group when compared with chamber controls. Statistically significant increases in several skeletal variations were observed in the high dose group when compared with chamber controls on both a per fetus and per litter basis. These observations included misshapen or bifid sternbrae, rudimentary cervical ribs, and well-formed cervical or lumbar ribs. There were statistically

significantly decreased incidences of an extra ossification site between the frontal bones of the skull in fetuses and litters of the mid- and high dose groups, of rudimentary cervical rib(s) in fetuses of the low dose group and rudimentary lumbar rib(s) in litters of the high dose group when compared with the chamber controls. However, decreased incidences of developmental variations were considered incidental findings and would not be considered adverse effects of treatment with the test material in any event.

Statistically significant increases in the incidences of total external, visceral, and skeletal malformations were observed in the high dose group when compared with the chamber controls on both a per fetus and per litter basis. A statistically significant decrease was observed in the incidence of total skeletal malformations in the low dose group when compared with chamber controls on both a per fetus and per litter basis. Individual malformations which were statistically significantly increased in the high dose group when compared with controls on both a per fetus and per litter basis included cleft palate, microtia, low set ears, multiple skull bones malformed and fusion of vertebral arches. Imperforate anus, ectrodactyly, malformed squamosal bones, fusion of the exoccipital and basioccipital bones, agenesis of the sternbrae and branched vertebral arches were statistically significantly increased in the high dose group when compared with chamber controls on a per fetus basis. A single, statistically significant reduction in the incidence of branched vertebral arches occurred in the low dose group when compared with chamber controls on both a per fetus and per litter basis. Additional malformations occurred in single or low incidences primarily in the high dose group, although tail malformations occurred in low incidences throughout the groups, including chamber controls. Individual external and visceral variations and malformations are presented in Appendix G and individual skeletal variations and malformations are presented in Appendix H.

3. FETAL SKELETAL OSSIFICATION SITES

Mean ossification site counts are presented in Table 9. There were no statistically significant differences in mean ossification sites between untreated controls and chamber controls.

There were statistically significant decreases in mean ossification sites of the mid phalanges of the forepaw and proximal phalanges, mid phalanges, and calcaneus of the hindpaw in high dose fetuses compared with chamber control fetuses. However, the aforementioned means were within the range considered normal for mouse fetuses at this stage of development. A statistically significant increase in mean rib

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ossification sites also was observed in the high dose fetuses when compared with chamber controls. Individual counts of ossification sites and litter averages are presented in Appendix I.

DISCUSSION

Maternal toxicity was observed at a dose of 3000 ppm, as indicated by moderate-to-marked maternal body weight gain suppression and reduced food consumption compared with controls. In addition, treatment-related reductions were observed in the high dose group for the mean number of live fetuses per litter and treatment-related increases were observed in the high dose group for the mean number of resorptions per litter, mean number of malformed fetuses per litter, and mean number of affected fetuses per litter. The reduction in live litter size contributed to a reduction in the mean number of fetuses/implantations observed at 3000 ppm compared with controls, while the increase in resorptions produced treatment-related increases in the mean number of resorptions/implantations and post-implantation loss. Similarly, the observed increase in the number of affected fetuses per litter was due to an increase in malformed fetuses in the high dose group compared with controls. No treatment-related clinical signs of toxicity or necropsy findings were observed during the study. In addition, chamber control animals were similar to untreated control animals for all parameters evaluated during this study.

While maternal food consumption was reduced slightly in animals exposed to 1000 ppm compared with controls for the GD 6-15, GD 9-12, and GD 0-18 intervals ($p < 0.05$), there were no corresponding reductions in maternal body weight or weight gain during these intervals or other evidence of maternal toxicity observed in mid-dose animals. Accordingly, the observed reductions in food consumption in mid-dose dams were considered incidental and not biologically meaningful. Similarly, the single statistically significant reduction in maternal food consumption for the GD 6-9 interval observed in mice exposed to 300 ppm was considered incidental and not biologically meaningful in the absence of similar reductions in food consumption at other intervals during the study or in the absence of a corresponding reduction in maternal body weight at this exposure level. In addition, maternal food consumption during the GD 6-9 interval was similar between untreated control mice and those exposed to 300 ppm, thereby lending additional support for the conclusion that the statistically significant reduction in maternal food consumption observed during this interval was not related to treatment with the test material.

Fetal toxicity was observed at a dose of 3000 ppm, as indicated by reduced fetal body weights and increased incidences of stunted fetuses (i.e., < 1.0 g) and of external, visceral, and skeletal abnormalities compared with controls. The overall incidence of skeletal variations was increased in the 3000 ppm dose group compared with that of controls, which

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was due primarily to increased incidences of misshapen or bifid sternbrae and rudimentary or supernumerary (well-formed) cervical and/or lumbar ribs. While increased incidences of fleshy tabs on the side of the mouth, misshapen squamosal bone, and misshapen pubis were observed in the high dose group compared with controls, these findings were only statistically significant when analyzed on a per fetus basis. Since the litter is the accepted unit of measure in the evaluation of developmental toxicity data, increased incidences of these fetal-based parameters were not considered biologically meaningful in the absence of statistical significance in the corresponding litter incidences. While high dose fetal body weights were reduced compared with those of controls ($p < 0.01$), the increased incidences of developmental variations observed in this study were not necessarily indicative of developmental delay.

Reductions in the fetal and litter-based incidences of abnormal hindlimb flexure in all treated groups or extra ossification sites between the frontal bones of the skull in mid- and high dose groups compared with controls were considered incidental findings in the absence of similar reductions in the incidences of other fetal malformations or developmental variations. Similarly, reductions in the fetal and litter-based incidences of overall external variations in all treated groups compared with controls were considered incidental findings since abnormal hindlimb flexure was the primary component of these parameters. In addition, the apparent reduction in the litter incidence of rudimentary lumbar ribs observed in the high dose group compared with controls ($p < 0.05$) appeared to be related to a corresponding increase in the fetal and litter incidences of supernumerary lumbar ribs in the 3000 ppm group rather than due to beneficial effects of the test material per se. Apparent reductions in the fetal incidences of asymmetric sternbrae and rudimentary cervical ribs in low dose animals compared with controls were considered incidental findings in the absence of similar reductions in the mid- and high dose groups and in the absence of statistically significant reductions in the litter incidences of these parameters.

On the other hand, treatment-related increases were observed in the high dose group for overall fetal and litter incidences of external, visceral, and skeletal malformations compared with controls. These findings were due primarily to increased fetal and litter incidences of cleft palate, microtia, low set ears, multiple skull bones malformed, and fused vertebral arches. Increased fetal incidences of microphthalmia, imperforate anus, ectrodactyly, malformed squamosal bone, fusion of exoccipital and basioccipital bones, sternbral agenesis, and branched vertebral arches in the high dose group compared with controls were not considered treatment-related in the absence of corresponding statistically significant

increases in the litter incidences of these parameters. Apparent reductions in the fetal and litter incidences of overall skeletal malformations and branched vertebral arches in low dose animals compared with controls ($p < 0.05$) were considered incidental findings in the absence of similar reductions in the mid- and high dose groups.

The mean number of fetal ossification sites was similar between treated and control groups for most parameters examined. However, a treatment-related increase in the number of rib ossification sites was observed in the high dose group compared with controls ($p < 0.01$) and was associated with increased incidences of rudimentary and supernumerary ribs. The reduced number of fetal ossification sites of proximal and mid phalanges of the hindpaw or of the calcaneus which were observed in the 3000 ppm group compared with controls ($p < 0.01$) may have been associated with reduced fetal body weights observed in that group. However, the observed values were greater than the expected normal ranges of 0-3, 0, and 0 ossification sites, respectively, which are used by this laboratory. Accordingly, the relationship of the observed reduction in ossification of proximal and mid phalanges and the calcaneus to developmental delay was not clear in the high dose group.

The absence of statistically significant differences between the chamber control and untreated control groups for fetal body weight and litter incidences of fetal malformations or developmental variations indicated that maternal stress was minimal during this study. Stress has been shown to induce malformations in mice (e.g., cleft palate and cranial malformations) [Greene and Kochhar, 1975; Harris et al., 1980; Hamm et al., 1977; Miller, 1972; Rosenzweig and Blaustein, 1970]. Therefore, stress could have presented a confounding variable in the interpretation of data obtained from a developmental toxicity study of the test material in mice using the inhalation route. However, maternal toxicity has been shown by Khara (1984) to produce a number of abnormalities which occurred in characteristic patterns in fetal mice, including cleft palate, exencephaly, fused vertebral arches, and supernumerary ribs. Thus, treatment-related malformations and developmental variations observed at 3000 ppm in the present study may have been exacerbated by concurrent maternal toxicity at this exposure level.

In conclusion, maternal toxicity was observed at a dose level of 3000 ppm, as indicated by maternal body weight gain suppression and reduced food consumption compared with controls. Accordingly, the maternal NOAEL (No Observable Adverse Effect Level) was established at 1000 ppm of dimethylcarbonate (MRD-91-073) under the conditions of this study. Fetal body weights were reduced in the high dose group

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compared with controls. In addition, there was an increased incidence of fetal malformations and developmental variations in the high dose group compared with chamber controls. Therefore, the fetal NOAEL was established at 1000 ppm of dimethylcarbonate under the conditions of this study. Teratogenic effects characterized by cleft palate, microtia, low set ears, multiple skull bone malformations, and fused vertebral arches were observed at a maternally-toxic exposure level of 3000 ppm when administered under the conditions of this study.

REFERENCES

- Bradley, J.V., Distribution Free Statistical Tests, Prentice Hall, Englewood Cliffs, N.J. 1968: Fischer Exact Test, p. 195-203
- Dixon and Massey, Introduction to Statistical Analysis, 3rd ed. McGraw-Hill Inc., New York (1969): Smith-Satterthwaite t-test, p. 114-116.
- Dunnett, C. "New Tables for Multiple Comparisons With A Control", Biometrics, Vol 20, 3, Sept. 1964: Dunnett's Test, p. 482-491.
- Greene, R. M., and Kochhar, D. M. (1975). Some aspects of corticosteroid-induced cleft palate: a review. Teratology 11, 47-56.
- Harris, S. B., Szczech, G. M., Stuckhardt, J. L., Riley, K., Purmalis, B. P., and Brunden, M. (1980). Evaluation of the Upj:TUC(ICR) strain of mice for use in teratology tests. J. Toxicol. Environ. Health 6, 155-165.
- Hemm, R. D., Arslanoglou, L., and Pollock, J. J. (1977). Cleft palate following prenatal food restriction in mice: association with elevated maternal corticosteroids. Teratology 15, 243-248.
- Hollander, M. and Wolfe, D. A., Nonparametric Statistical Methods, John Wiley and Sons, New York, 1973: Dunn's Summed Rank Test, p. 131; Kruskal-Wallis Test, p. 114-116; Jonckheere's Test, p. 120-123.
- Khera, K. S. (1984) Maternal toxicity- a possible factor in fetal malformations in mice. Teratology 29, 411-416.
- Miller, T. J. (1972). Cleft palate formation: the effects of fasting and iodoacetic acid on mice. Teratology 7, 177-182.
- Rosenzweig, S., and Blaustein, F. M. (1970). Cleft palate in A/J mice resulting from restraint and deprivation of food and water. Teratology 3, 47-52.
- Snedecor and Cochran, Statistical Methods, 6th Edition, The Iowa State Press, Ames, Iowa 1971: Bartlett's Test, p. 296-298; ANOVA, p. 277-279; Regression Analysis for Trend, p. 149-152; Regression Analysis for Lack of Fit, p. 160-164; Least Significant Difference; Chi-Square Test; Armitage's Test.

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PROTOCOL EXCEPTIONS

PROTOCOL DEVIATIONS:

CHAMBER HOMOGENEITY: The protocol stated that during trials for this study, homogeneity of test material distribution would be ascertained. The chamber concentration homogeneity conducted during the rangefinding study (107333) was determined to be acceptable for the Group 3 chamber for this study and, therefore, was not conducted during the trials for this study. This deviation did not adversely affect the study results or integrity.

ROOM NUMBER: The protocol states that the study animals were to have been housed in Room 505. Due to a typographical error in Protocol Change no. 1, the room number was listed as 509. During the acclimation period, animals were housed in Room 522 and subsequently moved to Room 505 on November 13, 1991, prior to cohabitation. Study animals remained in Room 505 for the remainder of the study. The receipt of animals into Room 522 and subsequent housing in Room 505 were protocol deviations. These deviations did not adversely affect the study results or integrity.

SINGLE HOUSING OF ANIMALS: The protocol states that the study animals were to have been single housed, except during the first week of quarantine and during mating. Animals were single housed after one day of quarantine. This deviation did not adversely affect the study results or integrity.

BODY WEIGHTS: Body weights inadvertently were not recorded during mating. This deviation did not adversely affect the study results or integrity.

WEIGHT: On gestation Day 0, several females weighed slightly less than 28 grams as specified by protocol. This deviation did not adversely affect the study results or integrity.

SHORTENED EXPOSURE DURATION: On November 24, 1991, the following animals which were due for initial exposures on GD 6 inadvertently were not loaded into the inhalation chambers for exposure starting at 08:45:

	0 ppm	300 ppm	1000 ppm	3000 ppm
Animal	JAB880	JAB846	JAB873	JAB902
Number	JAB923	JAB914	JAB784	JAB946
	JAB895	JAB952	JAB889	JAB926
		JAB829	JAB964	JAB936

Upon discovery of this oversight, exposures were stopped and the chambers were allowed to clear for 30 minutes. The

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fifteen overlooked animals then were added to the chambers. Exposures were resumed at 10:40 and continued until 15:15 so that the original group of animals received their scheduled 6-hour duration. As a result, animals that were overlooked were exposed for approximately 4.5 hours instead of the 6 hours specified by protocol. In addition, the original group of animals were in the exposure chambers for approximately 7 hours due to two sets of chamber equilibrations required rather than the usual one. After examination of the data for these animals, it was determined that this deviation did not adversely affect the study results or integrity of the study.

ACTUAL CHAMBER CONCENTRATION: On November 29, 1992, only 2 concentration samples were taken during exposures due to an equipment malfunction, instead of the 6 required by protocol. This deviation did not adversely affect the study results or integrity.

DELAYED EXPOSURES: Standard practice in developmental toxicity testing requires exposures to begin in the morning. Due to mechanical malfunction, exposures were delayed on December 7, 1991 until approximately 15:50. During this delay, animals were returned to the Room 505 and water lines were connected to the inhalation racks in an attempt to minimize any stress on the animals. After completion of repairs, all animals received the required 6 hours of exposure plus time for equilibration and had sufficient recovery time prior to exposures on December 8, 1991. This delay in exposures did not adversely affect the study results or integrity.

DISCONNECTED WATER LINES: During the morning room check on December 3, 1991, the water lines to the animal racks housing Groups 2 and 4 (300 ppm and 3000 ppm, respectively) were found to be disconnected. Due to technician oversight, the afternoon room check on December 2, 1991 had not been performed and the last confirmation of connected water lines was during the morning of December 2, 1991. Therefore, potential deprivation of water did not exceed 24 hours. Examination of body weight and food consumption data revealed that possible adverse effects were limited to four Group 2 females which displayed body weight gain suppression compared with other females in Group 2. All remaining Group 2 animals and all Group 4 animals did not appear to have been affected by the potential water deprivation. Cage papers were examined following discovery of the disconnected water lines and there were no remarkable differences in quantities of excreta, either within or between groups, thus providing additional evidence of a minimal effect on Group 2 and 4 animals.

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ENVIRONMENTAL DEVIATIONS:

TEMPERATURE: On several occasions, the temperature in the animal room was out of the range specified by protocol (68-76°F):

<u>Date</u>	<u>Temperature</u>
October 29, 1991	77°F
November 26, 1991	67°F

HUMIDITY: On several occasions, the humidity in the animal room was out of the range specified by protocol (40-70%RH):

<u>Date</u>	<u>Relative Humidity</u>
October 30, 1991	36%
November 5, 1991	32%
November 6, 1991	32%
November 7, 1991	32%

EXPOSURE CHAMBERS: On most occasions, the temperature in the exposure chambers was slightly higher (maximum 78°F) than the range specified by protocol (68-76°F). On several occasions, the relative humidity in the exposure chambers was slightly lower (minimum 24%) or slightly higher (maximum 73%) than the range specified by protocol (40-60%). These deviations did not adversely affect the study results or integrity.

No other circumstances occurred that would have affected the quality or integrity of the data.

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TABLE 1 - INCIDENCE OF GESTATION OBSERVATIONS (CONT.)

	TIME - G									
	0	1	2	3	4	5	6	7	8	9
GREEN THICK MATERIAL AROUND VAGINA										
0 PPM (UNTREATED CONTROL)	0	0	0	0	0	0	0	0	0	0
0 PPM (CHAMBER CONTROL)	0	0	0	0	0	0	0	0	0	0
300 PPM	0	0	0	0	0	0	0	0	0	0
1000 PPM	0	0	0	0	0	0	0	0	0	0
3000 PPM	0	0	0	0	0	0	0	0	0	0
YELLOW MATERIAL VAGINA										
0 PPM (UNTREATED CONTROL)	0	0	0	0	0	0	0	0	0	0
0 PPM (CHAMBER CONTROL)	0	0	0	0	0	0	0	0	0	0
300 PPM	0	0	0	0	0	0	0	0	0	0
1000 PPM	0	0	0	0	0	0	0	0	0	0
3000 PPM	0	0	0	0	0	0	0	0	0	0
AMENITIAL STAINING										
0 PPM (UNTREATED CONTROL)	0	0	0	0	0	0	0	0	0	0
0 PPM (CHAMBER CONTROL)	0	0	0	0	0	0	0	0	0	0
300 PPM	0	0	0	0	0	0	0	0	0	0
1000 PPM	0	0	0	0	0	0	0	0	0	0
3000 PPM	0	0	0	0	0	0	0	0	0	0
BROKEN LEG										
0 PPM (UNTREATED CONTROL)	0	0	0	0	0	0	0	0	0	0
0 PPM (CHAMBER CONTROL)	0	0	0	0	0	0	0	0	0	0
300 PPM	0	0	0	0	0	0	0	0	0	0
1000 PPM	0	0	0	0	0	0	0	0	0	0
3000 PPM	0	0	0	0	0	0	0	0	0	0
ANIMAL EUTHANIZED										
0 PPM (UNTREATED CONTROL)	0	0	0	0	0	0	0	0	0	0
0 PPM (CHAMBER CONTROL)	0	0	0	0	0	0	0	0	0	0
300 PPM	0	0	0	0	0	0	0	0	0	0
1000 PPM	0	0	0	0	0	0	0	0	0	0
3000 PPM	0	0	0	0	0	0	0	0	0	0
ANIMAL DELIVERED										
0 PPM (UNTREATED CONTROL)	0	0	0	0	0	0	0	0	0	0
0 PPM (CHAMBER CONTROL)	0	0	0	0	0	0	0	0	0	0
300 PPM	0	0	0	0	0	0	0	0	0	0
1000 PPM	0	0	0	0	0	0	0	0	0	0
3000 PPM	0	0	0	0	0	0	0	0	0	0

GD - GESTATION DAY

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KEY A - KEY TO STATISTICAL SYMBOLS

<u>No difference</u>	<u>p<0.05</u>	<u>p<0.01</u>	<u>Statistical Statement</u>
(PARAMETRIC)			
A-			No statistical difference among the means
	A	A+	Significant difference among the means
L-			No linear response to the dose levels
	L	L+	Response is linearly related to dose
	Q	Q+	Linear response shows lack of fit
	*	**	Mean significantly different from control mean
(NONPARAMETRIC)			
K-			No statistical difference among the means
	K	K+	Means differ significantly
J-			No ordered response to the dose levels
	J	J+	An ordered response to the dose levels
	*	**	Mean significantly different from control mean
t-test			
T-			No statistical difference among the means
	T	T+	Significant difference among the means
NT			Data not tested

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TABLE 2 - MEAN GESTATION BODY WEIGHTS AND BODY WEIGHT CHANGE (GRAMS)
(SEE KEY A FOR STATISTICAL ABBREVIATIONS)

	0 PPM (UNTREATED CONTROL)		0 PPM (CHAMBER CONTROL)		0 PPM (CHAMBER CONTROL)		300 PPM		1000 PPM		3000 PPM	
	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.
	28.48	1.30	28.58	1.36	28.58	1.36	29.24	1.82	28.63	1.56	28.78	1.72
	31.81	1.42	32.47	1.90	32.47	1.90	30.71	1.86	30.55	1.64	30.59	2.17
	36.66	1.87	36.90	1.95	36.90	1.95	43.03	3.66	42.80	2.12	37.09	3.05
	42.61	2.29	43.47	2.60	43.47	2.60	43.03	3.66	42.80	2.54	39.23	3.55
	51.14	3.16	51.92	3.40	51.92	3.40	51.23	4.69	51.67	3.11	45.92	4.90
	17.10	2.64	18.07	2.16	18.07	2.16	17.72	3.10	17.66	2.29	14.50	3.80
	34.04	1.96	33.85	1.97	33.85	1.97	33.51	2.89	34.00	2.11	31.42	2.43
	1.49	0.93	2.24	1.35	2.24	1.35	1.47	1.67	1.92	1.83	1.81	1.57
	0.82	0.31	1.65	1.31	1.65	1.31	1.75	1.35	1.62	0.94	1.13	1.53
	4.85	0.97	4.43	0.95	4.43	0.95	4.64	1.43	4.60	0.98	5.38	1.77
	0.97	0.31	0.82	0.32	0.82	0.32	0.98	0.31	0.94	0.30	1.48	1.43
	1.10	0.31	1.10	0.32	1.10	0.32	1.10	0.31	1.10	0.30	1.10	0.30
	5.95	0.97	6.57	1.04	6.57	1.04	6.57	1.04	6.03	0.98	6.03	1.48
	1.10	0.31	1.10	0.32	1.10	0.32	1.10	0.31	1.10	0.30	1.10	0.30

GD = Gestation Day
GD 18C = GD 18 body weight - Uterine Weight

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TABLE 2 - MEAN GESTATION BODY WEIGHTS AND BODY WEIGHT CHANGE (GRAMS) (CONT.)
(SEE KEY A FOR STATISTICAL ABBREVIATIONS)

	FEMALE		M		F		L		C	
	MEAN	STD. DEV.	MEAN	STD. DEV.	MEAN	STD. DEV.	MEAN	STD. DEV.	MEAN	STD. DEV.
0 PPM (UNTREATED CONTROL)	8.65	1.30	12.64	1.78	22.70	3.08	5.60	1.48	30	30
0 PPM (CHAMBER CONTROL)	8.40	1.10	12.65	1.62	23.25	3.11	5.18	1.84	30	30
300 PPM	8.45	3.05	12.32	3.22	22.03	4.36	.31	2.53	30	30
1000 PPM	8.87	1.84	12.25	2.06	23.04	2.88	5.37	2.14	30	30
3000 PPM	6.69	1.94	8.92	3.86	17.25	4.62	2.74	2.73	30	30

GD = Gestation Day
GD 18C = GD 18 body weight - Uterine Weight

ROW 1

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334

TABLE 3 - MEAN GESTATION FOOD CONSUMPTION (GRAMS)
(SEE KEY A FOR STATISTICAL ABBREVIATIONS)

	T-		T-		T-		T-		T-		T-	
	G	D	G	D	G	D	G	D	G	D	G	D
FEMALE												
0 PPM (UNTREATED CONTROL)												
MEAN	32.6	20.3	21.1	22.3	23.8	64.8	123.8					
STD.DEV.	3.1	3.2	7.0	3.4	2.5	9.5	17.0					
(N)	14	14	15	22	27	9	5					
0 PPM (CHAMBER CONTROL)												
MEAN	37.2	22.3	21.8	23.7	24.3	63.4	124.3					
STD.DEV.	7.7	7.2	3.0	5.1	2.3	5.0	4.5					
(N)	14	23	20	25	26	16	6					
	A-L-	AL	A+L+	K+J+	A+L+	A+L+	A+L+					
0 PPM (CHAMBER CONTROL)												
MEAN	37.2	22.3	21.8	23.7	24.3	63.4	124.3					
STD.DEV.	7.7	7.2	3.0	5.1	2.3	5.0	4.5					
(N)	14	23	20	25	26	16	6					
	A-L-	AL	A+L+	K+J+	A+L+	A+L+	A+L+					
300 PPM												
MEAN	35.0	17.6	19.9	21.6	24.0	60.1	118.3					
STD.DEV.	5.6	3.6	2.5	3.1	3.0	5.5	11.8					
(N)	14	14	19	25	24	12	6					
	A-L-	AL	A+L+	K+J+	A+L+	A+L+	A+L+					
1000 PPM												
MEAN	32.0	19.3	19.2	22.2	23.4	57.4	112.9					
STD.DEV.	3.8	4.5	1.9	2.6	3.1	5.6	6.9					
(N)	17	19	14	26	25	12	8					
	A-L-	AL	A+L+	K+J+	A+L+	A+L+	A+L+					
3000 PPM												
MEAN	34.2	17.1	14.9	17.8	20.8	48.0	100.7					
STD.DEV.	3.4	4.2	3.3	3.5	2.7	7.1	3.5					
(N)	6	15	21	28	30	9	3					
	A-L-	AL	A+L+	K+J+	A+L+	A+L+	A+L+					

GD = Gestation Day

RUN 1

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
 WITH DIMETHYLCARBONATE (MSD-91-073); 107334

TABLE 4 - INCIDENCE OF MATERNAL NECROPSY OBSERVATIONS

TERMINAL EUTHANASIA (N) -	0 FPM	300 FPM	1000 FPM	3000 FPM
(UNTREATED CONTROL) (CHAMBER CONTROL)	31	31	30	31
OBSERVATION:				
NO OBSERVABLE ABNORMALITIES	29	31	30	31
OVARY(IHS) -Surrounded by fluid-filled sec(s)	0	0	0	0
EYE -Damped (Right)	1	0	0	0
TAIL -Neurotic tip (5%)	1	0	0	0
POSTERIOR EUTHANASIA (N) -	0	0	0	1
HINDLEGS (Left): Extremely swollen	0	0	0	1
UTERUS: No evidence of implantation sites *	4	2	2	3

N = Number of pregnant animals examined

* = See Individual Maternal Necropsy Observations for postmortem data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
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TABLE 5 - MEAN UTERINE IMPLANTATION DATA
(SEE KEY A FOR STATISTICAL ABBREVIATIONS)

FEMALE	T-		T-		T-		T-		T-		T-		T-		T-		T-		T-			
	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.	MEAN	STD.DEV.		
0 PPM (UNTREATED CONTROL)	10.73	1.91	6.03	1.67	4.70	1.53	1.37	1.38	12.23	1.28	0.13	0.35	0.88	0.11	0.11	0.12	0.00	0.00	0.00	0.00	70.446767	8.903639
0 PPM (CHAMBER CONTROL)	11.67	1.79	6.43	2.47	5.23	2.10	0.80	1.03	12.50	1.63	0.03	0.18	0.93	0.06	0.08	0.08	0.00	0.00	0.00	0.00	75.461600	7.360641
0 PPM (CHAMBER CONTROL)	11.67	1.79	6.43	2.47	5.23	2.10	0.80	1.03	12.50	1.63	0.03	0.18	0.93	0.06	0.08	0.08	0.00	0.00	0.00	0.00	75.461600	7.360641
0 PPM (CHAMBER CONTROL)	11.67	1.79	6.43	2.47	5.23	2.10	0.80	1.03	12.50	1.63	0.03	0.18	0.93	0.06	0.08	0.08	0.00	0.00	0.00	0.00	75.461600	7.360641
300 PPM	11.20	2.31	5.20	1.81	6.03	1.94	0.90	1.06	12.20	2.19	0.07	0.25	0.92	0.08	0.09	0.09	0.00	0.00	0.00	0.00	73.926933	8.414005
1000 PPM	11.13	1.76	5.97	2.09	5.17	2.05	1.10	1.60	12.37	1.47	0.13	0.35	0.90	0.08	0.11	0.11	0.00	0.00	0.00	0.00	72.940467	9.082038
3000 PPM	9.23	3.40	4.57	2.24	4.67	1.83	2.80	3.12	12.50	1.48	0.47	1.85	0.75	0.22	0.24	0.24	0.00	0.00	0.00	0.00	61.222200	18.867403

TOT = Total; FET = Fetuses; FEM = Female; RESORP (RES) = Resorptions;
IMPLANTS (IMP) = Implantation Sites; F/I TRAN = Fetuses/Implantations Transformed

IMMUNIZATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MSD-91-073); 107334

TABLE 5 - MEAN UTERINE IMPLANTATION DATA (CONT.)
(SEE KEY A FOR STATISTICAL ABBREVIATIONS)

	R /	D /	I I	P P	M M	N O	N O	T T	T T	T T
UNITS>>>>>										
FEMALE										
0 PPM (UNFERTILIZED CONTROL)	18.523033	9.359	12.5	0.27	4.00	2.00	2.00			
MEAN	9.275156	2.859	11.2	0.52	2.00	2.00	2.00			
STD.DEV.	(N)	30	30	30	30	30	30			
0 PPM (CHAMBER CONTROL)	14.262500	6.478	6.6	0.63	4.00	1.00	1.00			
MEAN	7.103905	1.795	6.0	0.81	2.00	1.00	1.00			
STD.DEV.	(N)	30	30	30	30	30	30			
0 PPM (CHAMBER CONTROL)	14.262500	6.478	6.6	0.63	4.00	1.00	1.00			
MEAN	7.103905	1.795	6.0	0.81	2.00	1.00	1.00			
STD.DEV.	(N)	30	30	30	30	30	30			
300 PPM	15.392533	8.873	8.3	0.17	3.00	1.00	1.00			
MEAN	8.122657	2.024	9.3	0.38	2.00	1.00	1.00			
STD.DEV.	(N)	30	30	30	30	30	30			
1000 PPM	15.980233	9.430	9.6	0.43	3.00	2.00	2.00			
MEAN	8.661572	3.425	11.3	0.63	2.00	2.00	2.00			
STD.DEV.	(N)	30	30	30	30	30	30			
3000 PPM	26.252500	11.008	25.5	6.10	5.00	9.00	9.00			**
MEAN	17.037253	10.098	27.2	3.22	2.00	4.00	4.00			**
STD.DEV.	(N)	30	30	29	29	29	29			29

R/I TRAM = Resorptions/Implantations Transformed
D/I TRAM = Dead/Implantations Transformed
POST IMPLANTATION LOSS (Post I) = (Implantations - Live)/ Implantations
No. Mal. = Number of fetuses with malformations (external, visceral, or skeletal)
No. Var. = Number of fetuses with developmental variations (external, visceral, or skeletal)
No. Aff. = Number of fetuses affected (resorbed, malformed, or dead)

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334

TABLE 6 - MEAN FETAL BODY WEIGHTS (GRAMS)

DOSE	MALE		FEMALE	
	MEAN STD.DEV. (N)		MEAN STD.DEV. (N)	
0 PPM (UNTREATED CONTROL)	1.24 0.11 181		1.18 0.11 141	
0 PPM (CHAMBER CONTROL)	1.24 0.10 193		1.19 0.10 157	
0 PPM (CHAMBER CONTROL)	1.24 0.10 193		1.19 0.10 157	
300 PPM	1.27 0.12 154		1.19 0.12 181	
10 PPM	1.24 0.10 179		1.20 0.10 155	
3000 PPM	1.12 ** 0.14 137		1.07 ** 0.15 140	

* - Statistically significant difference from control (or between controls), p<0.05

** - Statistically significant difference from control (or between controls), p<0.01

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MSD-91-073): 107334

TABLE 7 - INCIDENCE OF FETAL OBSERVATIONS AND VARIATIONS

DOSE:	0 PPM (UNTREATED CONTROL)	0 PPM (CHAMBER CONTROL)
TOTAL FETUSES WITH EXTERNAL VARIATIONS	9/322 (7/30)	15/350 (11/30)
TOTAL LITTERS WITH EXTERNAL VARIATIONS		
TOTAL FETUSES WITH VISCERAL VARIATIONS	1/162 (1/30)	0/173 (0/30)
TOTAL LITTERS WITH VISCERAL VARIATIONS		
TOTAL FETUSES WITH SKELETAL VARIATIONS	106/160 (29/30)	106/177 (27/30)
TOTAL LITTERS WITH SKELETAL VARIATIONS		
EXTERNAL EXAMINATIONS		
-TOTAL FETUSES EXAMINED:	322 (30)	350 (30)
-TOTAL LITTERS EXAMINED:		
INDIVIDUAL EXTERNAL OBSERVATIONS		
ABNORMAL FLEXOR FIBLING(S)/HINDPAW(S) (DEFORMATION)	3 (2)	6 (5)
STUNTED (<1.0 gram)	5 (4)	7 (5)
TROPHOBLAST LARGER THAN NORMAL	0 (0)	1 (1)
INDIVIDUAL EXTERNAL VARIATIONS		
ABNORMAL FLEXOR FIBLING(S)/HINDPAW(S)	0 (6)	14 (11)
PROTRUDING TONGUE	1 (1)	0 (0)
HEAD/VISCERAL EXAMINATIONS		
-TOTAL FETUSES EXAMINED:	162 (30)	173 (30)
-TOTAL LITTERS EXAMINED:		
INDIVIDUAL VISCERAL VARIATIONS		
RENAL Pelves SLIGHTLY DILATED	1 (1)	0 (0)

NOTE: * p<0.05 by Fisher Exact Test
** p<0.01 by Fisher Exact Test
() Represents litter-based data

IMM: LATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-073): 107334

TABLE 7 - INCIDENCE OF FETAL OBSERVATIONS AND VARIATIONS (CONT.)

DOSE:	0 PPM (UNTREATED CONTROL)	0 PPM (CHAMBER CONTROL)
SKELETAL EXAMINATIONS	160 [30]	177 [30]
-TOTAL FETUSES EXAMINED:		
-TOTAL LITTERS EXAMINED:		
INDIVIDUAL SKELETAL VARIATIONS BETWEEN FRONTALS: SITE OF OSSIFICATION *	11 [7]	23 [11]
HYOID: HYPOPLASTIC	3 [1]	0 [0]
SUPRAOCCIPITAL: BIFID	1 [1]	0 [0]
BETWEEN STERNEBRAE: SITE OF OSSIFICATION	3 [3]	0 [0]
STERNEBRAE: MISSHAPEN	1 [1]	1 [1]
STERNEBRAE: BIFID	4 [4]	2 [2]
STERNEBRAE: ASYMMETRIC FORM	9 [7]	15 [10]
CERVICAL RIB(S): RUDIMENTARY	24 [14]	26 [13]
LUMBAR RIB(S): RUDIMENTARY	36 [20]	52 [25]
LUMBAR RIB(S): WELL-FORMED *	48 [20]	38 [17]
LUMBAR RIB(S): MISSHAPEN	1 [1]	0 [0]
BETWEEN VERTEBRAL ARCHES: SITE OF OSSIFICATION	0 [0]	2 [2]
VERTEBRAL ARCH (ES): MISSHAPEN	1 [1]	0 [0]
SUPERNUMERARY PRESACRAL VERTEBRAE	0 [0]	2 [1]
HINDPAW (Digit no. 1): SITE OF OSSIFICATION	0 [0]	1 [1]

NOTE: * P<0.05 by Fisher Exact Test
** P<0.01 by Fisher Exact Test
{ } Represents litter-based data

TABLE 7 - INCIDENCE OF FETAL OBSERVATIONS AND VARIATIONS (CONT.)

DOSE:	0 PPM (CHAMBER CONTROL)	300 PPM	1000 PPM	3000 PPM
TOTAL FETUSES WITH EXTERNAL VARIATIONS	15/350 [11/30]	4/337 * [3/30] *	5/334 * [3/30] *	11/277 [6/29]
TOTAL LITTERS WITH EXTERNAL VARIATIONS				
TOTAL FETUSES WITH VISCERAL VARIATIONS	0/173 [0/30]	0/170 [0/30]	1/171 [1/30]	1/137 [1/28]
TOTAL LITTERS WITH VISCERAL VARIATIONS				
TOTAL FETUSES WITH SKELETAL VARIATIONS ++	106/177 [27/30]	95/167 [27/30]	98/163 [29/30]	135/146 ** [29/29]
TOTAL LITTERS WITH SKELETAL VARIATIONS				
EXTERNAL LAMINATIONS				
-TOTAL FETUSES EXAMINED:	350 [30]	337 [30]	334 [30]	277 [29]
-TOTAL LITTERS EXAMINED:				
INDIVIDUAL EXTERNAL OBSERVATIONS				
ABNORMAL PLACENTA BINDINGS (S) (DISPOSITIONS)	6 [5]	1 [1]	3 [2]	3 [2]
STOWED (<1.0 gram) ++				
++	7 [5]	15 [6]	7 [5]	55 ** [18] **
TROPHOBLAST LARGER THAN NORMAL				
	1 [1]	0 [0]	0 [0]	0 [0]
MISSHAPE CRANIUM (POSSIBLE DEFORMATION)				
	0 [0]	2 [1]	0 [0]	0 [0]
INDIVIDUAL EXTERNAL VARIATIONS				
ABNORMAL PLACENTA BINDINGS (S)	14 [11]	4 * [3] *	5 * [3] *	3 * [3] *
PROTRUDING TONGUE	0 [0]	0 [0]	0 [0]	2 [2]
FLASBY TAB (S): SIDE OF MOOFE	0 [0]	0 [0]	0 [0]	6 ** [1]

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 + Dose-response trend, p<0.05 by Armitage Test
 ++ Dose-response trend, p<0.01 by Armitage Test
 [] Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
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TABLE 7 - INCIDENCE OF FETAL OBSERVATIONS AND VARIATIONS (CONT.)

DOSE:	0 PPM (CHAMBER CONTROL)	300 PPM	1000 PPM	3000 PPM
<u>HEAD/VISCERAL EXAMINATIONS</u>				
-TOTAL FETUSES EXAMINED:	173 [30]	170 [30]	171 [30]	137 [28]
-TOTAL LITTERS EXAMINED:				
<u>INDIVIDUAL VISCERAL OBSERVATIONS</u>				
KIDNEY(S) HEMORRHAGIC	0 [0]	2 [2]	1 [1]	0 [0]
<u>INDIVIDUAL VISCERAL VARIATIONS</u>				
DILATED RENAL PELLVIS	0 [0]	0 [0]	1 [1]	0 [0]
RIGHT ATRIUM AND VENTRICLE LARGER THAN NORMAL	0 [0]	0 [0]	0 [0]	1 [1]
<u>SKELETAL EXAMINATIONS</u>				
-TOTAL FETUSES EXAMINED:	177 [30]	167 [30]	163 [30]	140 [29]
-TOTAL LITTERS EXAMINED:				
<u>INDIVIDUAL SKELETAL OBSERVATIONS</u>				
BETWEEN FRONTALS: SITE OF OSSIFICATION ++	23 [11]	14 [9]	3 ** [2] **	3 ** [3] *
HYOID: HYPOPLASTIC	0 [0]	2 [1]	0 [0]	0 [0]
SQUAMOSAL: MISSHAPEN	0 [0]	0 [0]	0 [0]	7 ** [3]
BETWEEN STERNEBRAE: SITE OF OSSIFICATION	0 [0]	3 [2]	2 [1]	3 [2]
STERNEBRAE: MULTIPLE BONES HYPOPLASTIC	0 [0]	0 [0]	0 [0]	1 [1]
STERNEBRAE: MISSHAPEN	1 [1]	2 [2]	2 [2]	9 ** [7] *

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 + Dose-response trend, p<0.05 by Armitage Test
 ++ Dose-response trend, p<0.01 by Armitage Test
 [] Represents litter-based data

TABLE 7 - INCIDENCE OF FETAL OBSERVATIONS AND VARIATIONS (CONT.)

DOGS:	0 PPM (CHAMBER CONTROL)	300 PPM	1000 PPM	3000 PPM
INDIVIDUAL SKELETAL OBSERVATIONS (FEMURALS; RIBS)				
STERNUMS: ASYMMETRIC FORM ++	2 [2]	2 [2]	3 [3]	13 ** [11] **
CERVICAL RIB(S): MODERATELY ++	15 [10]	6 * [6]	6 [6]	20 [12]
CERVICAL RIB(S): WELL-FORMED ++	26 [13]	14 * [7]	18 [11]	73 ** [29] **
LUMBAR RIB(S): MODERATELY +	0 [0]	0 [0]	1 [1]	20 ** [11] **
LUMBAR RIB(S): WELL-FORMED +	52 [25]	44 [23]	42 [23]	29 [17] *
LUMBAR RIB(S): ASYMMETRIC FORM	36 [17]	39 [19]	43 [17]	85 ** [24] *
VERTEBRAS: MULTIPLE BONES HYPOPLASTIC	0 [0]	1 [1]	0 [0]	0 [0]
BETWEEN OR CRANIAL TO VERTEBRAL ARCHES: SITE OF OSSIFICATION	0 [0]	0 [0]	0 [0]	1 [1]
VERTEBRAL ARCH(ES): MISSHAPEN	2 [2]	1 [1]	1 [1]	3 [3]
SOVEREIGNARY PRESACRAL VERTEBRAS	0 [0]	0 [0]	1 [1]	1 [1]
FOURS: MISSHAPEN	2 [1]	4 [2]	0 [0]	5 [2]
SINGLES (Digit no. 1): SITE OF OSSIFICATION	0 [0]	0 [0]	0 [0]	4 * [2]
NOTE:	1 [1]	0 [0]	0 [0]	0 [0]

* p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 + Dose-response trend, p<0.05 by Armitage Test
 ++ Dose-response trend, p<0.01 by Armitage Test
 [] Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MRD-91-C/3); 107334

TABLE 6 - INCIDENCE OF FETAL MALFORMATIONS

DOSE:	0 PPM (UNTREATED CONTROL)	0 PPM (CHAMBER CONTROL)
TOTAL FETUSES WITH EXTERNAL MALFORMATIONS	2/322 (2/30)	7/350 (5/30)
TOTAL LITTERS WITH EXTERNAL MALFORMATIONS	0/322 (0/30)	2/173 (2/30)
TOTAL FETUSES WITH VISCERAL MALFORMATIONS	7/160 (6/30)	12/177 (10/30)
TOTAL LITTERS WITH VISCERAL MALFORMATIONS	322 (30)	350 (30)
TOTAL FETUSES WITH SKELETAL MALFORMATIONS	0 (0)	3 (2)
TOTAL LITTERS WITH SKELETAL MALFORMATIONS	0 (0)	1 (1)
INDIVIDUAL EXTERNAL MALFORMATIONS	1 (1)	2 (1)
-TOTAL FETUSES EXAMINED:	0 (0)	1 (1)
-TOTAL LITTERS EXAMINED:	1 (1)	0 (0)
CLEFT PALATE	1 (1)	0 (0)
POLYDACTYLY FOLEX HINDPAW	1 (1)	0 (0)
KINKED TAIL	0 (0)	0 (0)
BENT TAIL	1 (1)	0 (0)
ANENCEPHALY	1 (1)	0 (0)
MICROTIA	1 (1)	0 (0)
LOW SET EARS	1 (1)	0 (0)
BILATERAL ASLEPHARIA	1 (1)	0 (0)

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 [] Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYL CARBONATE (MWD-91-073); 107334

TABLE 8 - INCIDENCE OF FETAL MALFORMATIONS (CONT.)

DOSE:	0 PPM (UNTREATED CONTROL)	0 PPM (CLAMBER CONTROL)
<u>INDIVIDUAL VISCERAL MALFORMATIONS:</u>		
-TOTAL FETUSES EXAMINED:	142 [30]	173 [30]
-TOTAL LITTERS EXAMINED:	0 [0]	2 [2]
<u>HEAD: CLEFT PALATE</u>		
<u>INDIVIDUAL SKELETAL MALFORMATIONS:</u>		
-TOTAL FETUSES EXAMINED:	160 [30]	177 [30]
-TOTAL LITTERS EXAMINED:	1 [1]	0 [0]
<u>MULTIPLE SKULL BONES MALFORMED</u>		
HYOID: AGENESIS	1 [1]	0 [0]
PALATINES: MALFORMED	0 [0]	1 [1]
EXOCCIPITAL(S) AND BASIOCCIPITAL: FUSED	0 [0]	1 [1]
STERNURAE: FUSED WITH SITE OF OSSIFICATION	1 [1]	0 [0]
STERNURAE: AGENESIS	0 [0]	1 [1]
VERTERAL ARCH (ES): BRANCHED	4 [4]	11 [10]

NOTE: * p<0.05 by Fisher Exact Test
** p<0.01 by Fisher Exact Test
[] Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MEO-91-073): 107334

TABLE 0 - INCIDENCE OF FETAL MALFORMATIONS (CONT.)

DOSE:	0 PPM (CHAMBER CONTROL)	300 PPM	1000 PPM	3000 PPM
TOTAL FETUSES WITH EXTERNAL MALFORMATIONS ++	7/350	2/337	2/334	145/277 **
TOTAL LITTERS WITH EXTERNAL MALFORMATIONS ++	(5/30)	(2/30)	(2/30)	(26/29) **
TOTAL FETUSES WITH VISCERAL MALFORMATIONS ++	2/173	0/170	0/171	73/137 **
TOTAL LITTERS WITH VISCERAL MALFORMATIONS ++	(2/30)	(0/30)	(0/30)	(24/28) **
TOTAL FETUSES WITH SKELETAL MALFORMATIONS ++	12/177	3/167 *	12/163	100/140 **
TOTAL LITTERS WITH SKELETAL MALFORMATIONS ++	(10/30)	(3/30) *	(10/30)	(28/29) **
INDIVIDUAL EXTERNAL MALFORMATIONS				
-TOTAL FETUSES EXAMINED:	350	337	334	277
-TOTAL LITTERS EXAMINED:	(30)	(30)	(30)	(29)
CLEFT PALATE ++				
++	3	0	1	140 **
	(2)	(0)	(1)	(26) **
MICROTIA ++				
	0	0	0	24 **
	(0)	(0)	(0)	(5) *
LOW SET EAR(S)				
	0	0	0	13 **
	(0)	(0)	(0)	(5) *
ANENCEPHALY				
	0	0	0	1
	(0)	(0)	(0)	(1)
EXENCEPHALY				
	0	0	0	1
	(0)	(0)	(0)	(1)
DOMED HEAD				
	0	0	0	1
	(0)	(0)	(0)	(1)
EYE OPEN				
	0	0	0	1
	(0)	(0)	(0)	(1)
MICROPTHALMIA				
	0	0	0	1
	(0)	(0)	(0)	(1)
EXOPHTHALMOS				
	0	0	0	3
	(0)	(0)	(0)	(1)
MICROGNATHIA				
	0	0	0	1
	(0)	(0)	(0)	(1)
	0	0	0	2
	(0)	(0)	(0)	(1)

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 + Dose-response trend, p<0.05 by Armitage Test
 ++ Dose-response trend, p<0.01 by Armitage Test
 {} Represents litter-based data

IRADIATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MCD-91-073); 107334

TABLE 8 - INCIDENCE OF FETAL MALFORMATIONS (CONT.)

DOSE:	0 PPM	300 PPM	1000 PPM	3000 PPM
IMPERFORATE ANUS	0 [0]	0 [0]	0 [0]	5 [3]
GENITAL TUBERCLE SMALLER THAN NORMAL	0 [0]	0 [0]	0 [0]	3 [2]
POLYDACTILY FORELIMB	1 [1]	0 [0]	0 [0]	0 [0]
ECTRODACTILY	0 [0]	0 [0]	0 [0]	4 [2]
BRACHYDACTILY	0 [0]	0 [0]	0 [0]	3 [1]
SYNDACTILY	0 [0]	0 [0]	0 [0]	2 [2]
SHORT DIGIT(S); HINDPAW	0 [0]	0 [0]	0 [0]	1 [1]
SHORT TAIL	0 [0]	0 [0]	0 [0]	1 [1]
KINKED TAIL	2 [1]	0 [0]	1 [1]	4 [4]
BENT TAIL	1 [1]	2 [2]	0 [0]	3 [3]
INDIVIDUAL VISCERAL MALFORMATIONS: -TOTAL FETUSES EXAMINED: -TOTAL LITTERS EXAMINED:	173 [30]	170 [30]	171 [30]	137 [28]
HEAD: CLEFT PALATE ++	2 [2]	0 [0]	0 [0]	73 [24]
ECTOPIC OVARIES	0 [0]	0 [0]	0 [0]	3 [2]
ECTOPIC KIDNEYS	0 [0]	0 [0]	0 [0]	1 [1]

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 † Dose-response trend, p<0.05 by Armitage Test
 ‡ Dose-response trend, p<0.01 by Armitage Test
 [] Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MED-91-073): 107334

TABLE 6 - INCIDENCE OF FETAL MALFORMATIONS (CONT.)

DOSE:	0 PPM	300 PPM	1000 PPM	3000 PPM
INDIVIDUAL SKELETAL MALFORMATIONS:				
-TOTAL FETUSES EXAMINED:	177 [30]	167 [30]	163 [30]	140 [29]
-TOTAL LITTERS EXAMINED:	0 [0]	0 [0]	0 [0]	80 ** [22] **
MULTIPLE SKULL BONES MALFORMED ++	0 [0]	0 [0]	0 [0]	10 ** [4]
SQUAMOSAL: MALFORMED	0 [0]	0 [0]	0 [0]	0 [0]
PALATINES: MALFORMED	1 [1]	0 [0]	1 [1]	0 [0]
EXOCCIPITAL(S) AND BASIOCCIPITAL: FUSED	1 [1]	0 [0]	3 [3]	7 * [4]
TYMPANICS: MALFORMED	0 [0]	0 [0]	0 [0]	1 [1]
STERNERAE: MALFORMED	0 [0]	0 [0]	0 [0]	3 [3]
STERNERAE: FUSED	0 [0]	0 [0]	0 [0]	2 [1]
STERNERAE: AGENESIS	1 [1]	0 [0]	0 [0]	6 * [5]
STERNERAE: BRANCHED	0 [0]	0 [0]	1 [1]	0 [0]

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 † Dose-response trend, p<0.05 by Armitage Test
 ++ Dose-response trend, p<0.01 by Armitage Test
 [] Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE
WITH DIMETHYLCARBONATE (MWD-91-073); 107334

TABLE 8 - INCIDENCE OF FETAL MALFORMATIONS (CONT.)

DOSE:	0 PPM	300 PPM	1000 PPM	3000 PPM
INDIVIDUAL SKELETAL MALFORMATIONS (CONT.)				
CERVICAL RIB(S): BRANCHED	0 [0]	1 [1]	0 [0]	0 [0]
THORACIC RIB(S): FUSED	0 [0]	0 [0]	0 [0]	2 [2]
VERTEBRAL ARCH (ES): BRANCHED ++ +	11 [10]	2 [2]	7 [5]	20 [13]
VERTEBRAL ARCH (ES): MALFORMED	0 [0]	0 [0]	0 [0]	2 [1]
VERTEBRAL ARCHES: FUSED	0 [0]	0 [0]	0 [0]	6 [6]
FOREPAW: AGENESIS	0 [0]	0 [0]	0 [0]	3 [2]
FORELIMS (S)/HINDLIMS (S): SHORT	0 [0]	0 [0]	0 [0]	2 [1]
HINDPAW: AGENESIS	0 [0]	0 [0]	0 [0]	2 [1]
METATARSALS: FUSED	0 [0]	0 [0]	0 [0]	1 [1]

NOTE: * p<0.05 by Fisher Exact Test
 ** p<0.01 by Fisher Exact Test
 + Dose-response trend, p<0.05 by Armitage Test
 ++ Dose-response trend, p<0.01 by Armitage Test
 {} Represents litter-based data

INHALATION DEVELOPMENTAL TOXICITY STUDY IN MICE WITH DIMETHYLCARBONATE (MRD-91-073): 107334

TABLE 9 - MEAN SKELETAL OSSIFICATION SITES (SEE KEY A FOR STATISTICAL SYMBOLS)

	-F O R E P A W-			- S R I E -			V E R T E B R A E			H I M D P A W					
	M C	M P	D P	S T	R I	E (7)	L (6)	S (4)	T (13)	M T	P (0-3)	M P	D P	C C	T L
NORMAL RANGE:	(4)	(2)	(5)	(1-6)	(13 pes)	(7)	(6)	(4)	(13)	NT	(0-3)	(0)	(5)	(0)	(0)
0 PPM (UNTREATED CONTROL)	MEAN	4.0	4.0	6.0	13.5	7.0	6.0	4.0	13.0	NT	5.0	4.0	5.0	2.2	5.0
	STD.DEV.	0.0	0.0	0.1	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.6	0.6
	(N)	30	30	30	30	30	30	30	30	30	30	30	30	30	30
0 PPM (CHAMBER CONTROL)	MEAN	4.0	4.0	6.0	13.5	7.0	6.0	4.0	13.0	NT	5.0	4.0	5.0	2.0	5.0
	STD.DEV.	0.0	0.0	0.0	0.3	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	(N)	30	30	30	30	30	30	30	30	30	30	30	30	30	30
0 PPM (CHAMBER CONTROL)	MEAN	4.0	4.0	6.0	13.5	7.0	6.0	4.0	13.0	NT	5.0	4.0	5.0	2.0	5.0
	STD.DEV.	0.0	0.0	0.0	0.3	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	(N)	30	30	30	30	30	30	30	30	30	30	30	30	30	30
300 PPM	MEAN	4.0	4.0	5.0	13.5	7.0	6.0	4.0	13.0	NT	5.0	4.0	5.0	2.3	5.0
	STD.DEV.	0.0	0.1	0.6	0.3	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.5	0.7
	(N)	30	30	30	30	30	30	30	30	30	30	30	30	30	30
1000 PPM	MEAN	4.0	4.0	5.0	13.5	7.0	6.0	4.0	13.0	NT	5.0	4.0	5.0	2.1	5.0
	STD.DEV.	0.0	0.0	0.4	0.3	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1
	(N)	30	30	30	30	30	30	30	30	30	30	30	30	30	30
3000 PPM	MEAN	3.9	3.7	2.2	14.3	7.0	6.1	4.0	13.0	NT	5.0	4.0	5.0	2.1	5.0
	STD.DEV.	0.3	1.0	0.9	0.5	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.8	0.1
	(N)	29	29	29	29	29	29	29	29	29	29	29	29	29	29

MC - METACARPALS
 PP - PROXIMAL PHALANGES
 MP - MID PHALANGES
 DP - DISTAL PHALANGES
 ST - STERNEBRAE
 RI - RIBS
 CE - CERVICAL
 T - THORACIC
 L - LUMBAR
 S - SACRAL
 MT - METATARSALS
 CC - CALCANEUS
 TL - TALUS

KEY B - KEY TO INLIFE OBSERVATIONS ABBREVIATIONS

+ = Observation present

- = Observation not present at scheduled interval

