US EPA ARCHIVE DOCUMENT

BK-866
7831

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY

WASHINGTON, DC 20460

007831

MAR 23 1990

OFFICE OF
PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: p-Dichlorobenzene (PDCB): Review of Toxicology Studies Submitted by the Registrant in Response to the October 21, 1987 Data Call-In Notice

TOX Chem. No.: 632
Project No.: 9-1751 and 0-0531
Record No.: 247788 and 258232
MRID No.: 411088-01 and 413150-01

FROM: Elizabeth A. Doyle, Ph.D. E.A. Doyle 3/13/90
Review Section I
Toxicology Branch II (HFAS)
Health Effects Division (H7509C)

TO: Walter Francis, PM 32
Antimicrobial Program Branch
Registration Division (H7505C)

THRU: Yiannakis M. Ioannou, Ph.D., Section Head Y.M. Ioannou 3/13/
Review Section I
Toxicology Branch II (HFAS)
Health Effects Division (H7509C)

and

Marcia van Gemert, Ph.D., Chief M van Gemert 3/21/90
Toxicology Branch II (HFAS)
Health Effects Division (H7509C)

Registrant: Chemical Manufacturers Association

Toxicology Branch II has reviewed two studies submitted by the registrant in response to the October 21, 1987 Data Call-In Notice on p-dichlorobenzene:

1. A 21-Day Dermal Toxicity Study in Rats with Para-Dichlorobenzene

2. Paradichlorobenzene - Two-Generation Reproduction Study of Inhaled Paradichlorobenzene in Sprague-Dawley (CD) Rats

1 of 2

1. <u>21-Day Dermal Toxicity Study</u> (MRID No. 413150-01) - Five male and five female CD (Sprague-Dawley derived) rats were treated with 0, 75, 150 or 300 mg p-DCB/kg body weight/day. The test material was dissolved in light mineral oil and painted on the backs of the rats five days per week for three weeks. The test site was covered with an impermeable polyethylene patch for six hours. After six hours, the patch was removed and residual test material wiped away. No treatment related effects were reported. This study was classified **Core - Supplementary** due to insufficiently high treatment levels. Data at higher dose levels will be required to satisfy this data gap.

2. <u>Two-Generation Reproduction Study by the Inhalation Route</u> (MRID No. 411088-01) - Twenty-eight male and female Sprague-Dawley (CD) rats were exposed for ten weeks to the test material atmosphere and then bred to produce F_1 litters. Twenty-eight randomly selected F_1 pups/sex /group were exposed to the test atmosphere for eleven weeks and then bred to produce F_2 litters. Exposure was for six hours per day, seven days per week for ten or eleven weeks prior to breeding, for the first 19 days of gestation, and postnatal day 5 until weaning. Target test atmosphere concentrations were 0, 50, 150 and 450 ppm. Test atmosphere analyses were not adequately conducted and the concentration of the test atmosphere could not be accurately determined at any treatment level. Hyaline droplet nephrosis in males was observed at all treatment levels tested. High dose females had litters with reduced numbers of live pups, reduced pup weights and decreased pup survival at day 4 of lactation. This study was classified **Core - Supplementary** due to the inadequate test atmosphere sampling system and the lack of a systemic NOEL in male rats.

Reviewed by: Elizabeth A. Doyle, Ph.D. E.A. Doyle 2/2/90
Section I, Tox. Branch II (HFAS) (H7509C)
Secondary Reviewer: Yiannakis M. Ioannou, Ph.D., Section Head M.I. 2/2/9
Section I, Tox. Branch II (HFAS) (H7509C)

DATA EVALUATION REPORT 007831

STUDY TYPE: 21-Day Dermal - Rats (82-2) TOX. CHEM. NO.: ~~4800~~ 632

MRID NO.: 413150-01

TEST MATERIAL: Para-dichlorobenzene

STUDY NUMBER: 88-3384

SPONSOR: Chemical Manufacturers Association
2501 M St., NW
Washington, D.C. 20037

TESTING FACILITY: Bio/dynamics, Inc.
Mettlers Road
East Millstone, New Jersey 008875

TITLE OF REPORT: A 21-Day Dermal Toxicity Study in Rats with Para-Dichlorobenzene

AUTHOR(S): Carol S. Auletta

REPORT ISSUED: October 26, 1989

CONCLUSIONS: No treatment related effects were observed in this study due to dermal exposure of Sprague-Dawley rats to 75, 150 or 300 mg of p-dichlorobenzene/kg/day for six hours per day, five days per week for three consecutive weeks.

Classification: core - Supplementary
(Deficient in that the dose levels selected did not approximate the limit test and failed to elicit toxic signs.)

This study does not satisfy the guideline requirements (82-2) for a "21-Day Dermal Toxicity Study in Rats".

3

A. MATERIALS:

1. Test compound: para-dichlorobenzene Description: flakes
Batch: container #6, Purity: 100%, contaminants: none
(The purity was "documented by the sponsor."

2. Test animals: Species: rat, Strain: CD (Sprague-Dawley derived), Age: 11 to 14 weeks, Weight: Males - 405-484, Females - 249-295, Source: Charles River Breeding Laboratories, Inc., Kingston, NY

B. STUDY DESIGN:

1. Animal assignment - Animals were assigned randomly to the following test groups:

Test Group	Dose (mg/kg/day)	Main Study 3 weeks male	Main Study 3 weeks female	Interim Sac. - months male	Interim Sac. - months female
1 Control	0	5	5		
2 Low (LDT)	75	5	5		
3 Mid (MDT)	150	5	5		
4 High (HDT)	300	5	5		

2. Sample preparation - The active ingredient was dissolved in a light mineral oil vehicle (Fisher Scientific, Lot No. 871698) for application to the test animals. Prior to the initiation of the study, the stability of the test material was verified by the Metabolism and Analytical Chemistry Department of Bio/dynamics, Inc. to be greater than eight days. Analyses were conducted by gas chromatography of diluted test materials. Fresh test material was prepared weekly and the concentrations confirmed.

Results - The concentrations of the three batches of test material were found to be 106 ± 6%, 97 ± 6% and 97 ± 6% of nominal for the low, mid and high doses, respectively.

3. Treatment Application - Rats were individually housed in stainless steel wire mesh cages. Lighting was maintained on a 12 hour light and 12 hour dark schedule.

Prior to initiation of dosing, the hair on the back of each animal was removed with a clipper. Care was taken to avoid abrading the skin. Animals were reclipped as necessary during the study. The test material was prepared so that 2 ml/kg/dose was applied at each treatment. Rats were treated daily, five days per week for three weeks. The test material was spread uniformly over the test site (area not indicated). The test site was covered with a polyethylene patch (approximately 3 cm x 3 cm), which was then secured by an adhesive bandage wrapped around the trunk. Control animals were

4

treated in the same manner with vehicle only. The wrappings were removed after about 6 hours of exposure and residual material was gently wiped from the exposure site.

Results - No significant dermal irritation was reported. Some focal erythema and necrosis were reported in two control and three high dose animals, but these observations were attributed to the bandages.

4. Animals received food (Purina Certified Rodent ChowBrand Animal Diet #5002) and water ad libitum.

5. Statistics - The following procedures were utilized in analyzing the numerical data: Analysis of variance, Dunnett's test, Bartlett's test, Kruskal-Wallis test, summed rank test, regression analysis for trends and lack of fit and Jonckheere's statistic.

6. Quality assurance was documented by signed and dated quality assurance statement. Although listed in the table of contents, no GLP statement was included.

C. METHODS AND RESULTS:

1. Observations - Animals were inspected twice daily for signs of toxicity and mortality.

   Results - Toxicity - No treatment related signs of toxicity were reported.

   Mortality (survival) - All animals survived to termination.

2. Body weight - The animals were weighed twice prior to the initiation of the study, weekly for three weeks, and at termination.

   Results - No treatment related effects were reported.

3. Food consumption and compound intake - Consumption was weekly and mean daily food consumption was calculated.

   Results - Food consumption - No treatment related effects were reported.

4. Ophthalmological examinations were not performed.

5. Blood was collected before treatment and at termination for hematology and clinical analysis from all animals. The CHECKED (X) parameters were examined.

a. Hematology:

X		X	
X	Hematocrit (HCT)		Total plasma protein (TP)
X	Hemoglobin (HGB)	X	Leukocyte differential count
X	Leukocyte count (WBC)	X	Mean corpuscular HGB (MCH)
X	Erythrocyte count (RBC)	X	Mean corpuscular HGB conc. (MCHC)
X	Platelet count	X	Mean corpuscular volume (MCV)

Results - No significant treatment related effects were reported.

b. Clinical Chemistry

X		X	
	Electrolytes:		Other:
X	Calcium	X	Albumin
X	Chloride	X	Blood creatinine
	Magnesium	X	Blood urea nitrogen
X	Phosphorous		Cholesterol
X	Potassium		Globulins
X	Sodium	X	Glucose
	Enzymes:	X	Total Bilirubin
X	Alkaline phosphatase	X	Total Protein
	Cholinesterase	X	Triglycerides
	Creatinine phosphokinase		
	Lactic acid dehydrogenase		
X	Serum alanine aminotransferase (also SGPT)		
X	Serum aspartate aminotransferase (also SGOT)		

Results - No significant treatment related effects were reported.

6. Urinalysis - Not performed.

7. Sacrifice and Pathology - All rats on the study were subjected to gross pathological examination. Organ weights were taken for brain, liver, kidneys and testes with epididymides. Kidneys, liver, skin (treated and untreated) and gross lesions were fixed in 10% neutral buffered formalin. Samples from the controls and high dose group were subjected to histological examination.

Results

a. Organ weight - Kidney weights, kidney weight/body weight ratios and kidney weight/brain weight ratios in females demonstrated a slight dose related increase. This effect did not occur in males. The magnitude of the increases was small with no concomitant histopathological changes in this organ. Thus, this finding is of questionable biological significance.

b. Gross pathology - Two male and two female rats from the high dose group and two male and one female rats from the low dose group exhibited gross reddening of the mandibular lymph nodes. The report states that "these findings in mandibular lymph nodes in

rats are not uncommon and may be due to drainage from the turbinates ..." This observation does not appear to be treatment related as it did not occur in the mid dose group.

c. Microscopic pathology - The mandibular lymph nodes which were reddened exhibited plasma cell hyperplasia, congestion and/or erythrocytes in the sinuses upon microscropic examination.

D. DISCUSSION: No apparent treatment related effects were reported due to repeated exposure to 75, 150 or 300 mg p-dichlorobenzene/kg/day. The registrant cited a pilot study in which p-dichlorobenzene dose levels of 250, 500 and 1000 mg/kg/day were applied in an acetone vehicle. The acetone vehicle was found to be too irritating to continue, so the study was terminated after day 5. Mineral oil was chosen as an alternate vehicle. The solubility of the test material in the mineral oil was stated as the reason for the selection of the lower concentrations. No other attempts to find an alternate vehicle which was more compatible with the test material were cited.

Classification: core - Supplementary

(Deficient in that the dose levels selected did not approximate the limit test and failed to elicit toxic signs.)

This study does not satisfy the guideline requirements (82-2) for a "21-Day Dermal Toxicity Study in Rats".

7

Primary Review by: Elizabeth A. Doyle, Ph.D. E.A.D. 3/8/90
Review Section I, Tox Branch-HFAS/HED (H7509C)
Secondary Review by: Yiannakis M. Ioannou, Ph.D YMI 3/7/90
Section Head, Review Section I, Tox Branch-HFAS/HED (H7509C)

007831

DATA EVALUATION RECORD

STUDY TYPE: Two-Generation Reproduction (Inhalation) - Rat (Guideline 83-4)

TOX. CHEM. NO.: 632

MRID NO.: 411088-01

TEST MATERIAL: Paradichlorobenzene

SYNONYMS: PDCB

STUDY NUMBER: 86-81-90605

SPONSOR: Chlorobenzene Producers Association

TESTING FACILITY: Bushy Run Research Center
R. D. 4, Mellon Road
Export, Pennsylvania 15632

TITLE OF REPORT: Paradichlorobenzene - Two-Generation Reproduction Study of Inhaled Paradichlorobenzene in Sprague-Dawley (CD) Rats

AUTHORS: Rochelle W. Tyl, Ph.D., DABT
Teresa L. Neeper-Bradley, Ph.D.

DATE REPORT ISSUED: January 16, 1989

CONCLUSIONS: Paradichlorobenzene caused hyaline droplet nephrosis in male rats at concentrations ≥ 50 ppm in the test atmosphere. Exposure of pregnant female rats to atmospheres containing 450 ppm PDCB resulted in litters with reduced numbers of live pups, reduced pup weights and decreased pup survival at day 4 of lactation.

NOEL for Reproductive Toxicity = 150 ppm based on litters with reduced numbers of live pups, pup weights and 4-day survival
LOEL for Reproductive Toxicity = 450 ppm
NOEL for Systemic Effects < 50 ppm
LOEL for Systemic Effects = 50 ppm based on hyaline droplet nephrosis in male rats

Core Classification: Supplementary

This study does not satisfy the guideline requirements (83-4) for a "Two Generation Reproduction Study in Rats".

8

I. PROTOCOL

A. Materials

1. Test Material: paradichlorobenzene (CAS No. 106-47-7), Purity: 100%
Description: white crystalline solid or clear, aromatic liquid
Source: PPG Industries, New Martinsville, WV, Lot No. 987,
Drum No. A0507, A98704, "All-PCDB"

2. Test Species: Six week old male and female Sprague-Dawley (CD) rats from Charles River Breeding Laboratories, Kingston, NY.
Body weights: males - 180.9-226.0 g, females - 125.5-158.2 g

3. Diet: Certified Ground Rodent Chow, Agway, Inc., St. Mary's, OH
Food and water were available ad libitum except during exposure periods.

B. Procedures and Study Design

1. Atmosphere Generation: Per the study report, "Target exposure concentrations for the exposure chambers were 0.0, 50.0, 150.0 and 450.0 ppm. PDCB vapor was generated by metering in-house and filtered compressed air at a flow rate of 0.6-3.5 L/minute (depending upon the target concentration) into a heated (130-136°C) 12-liter stainless steel pot ... containing PDCB. The resulting vapor was introduced into the exposure chamber through a heated glass pyrex outlet tube (5/8" diam.) which joined the pot lid to the chamber intake line. The amount of PDCB in the pot, 1.8-2.8 kg, as well as the compressed airflow rate determined target chamber concentrations."

2. Atmosphere Analysis: Per the study report, "Gas chromatography with flame ionization detection (Perkin Elmer 8500) was used to measure PDCB concentrations in the exposure chamber atmospheres. Two vapor sampling techniques, one using gas-tight syringes and the other employing charcoal tube sampling methods, were used during the study. Direct syringe sampling from stainless steel tubes placed in the breathing zone of the animals was used during the first 82 exposure days. Direct syringe sampling from 3 inches inside the exposure chamber (no tubing) was employed for exposure days 83-171, and coconut-based charcoal sorbent tubes (placed within the breathing zone of the animals) were used for sampling during the remainder of the study (exposure days 172-284). At least six (6) samples were taken daily form each exposure chamber (including control chamber) for exposure days 1-171. For days 172-282, three (3) samples were taken daily from each exposure chamber atmosphere and one from control chamber atmospheres."

3. Exposure and Mating: "F_0 study animals were exposed for 10 weeks and then bred to produce F_1 litters. Twenty-eight (28) randomly selected F_1 pups/sex/group were exposed to the test chemical for eleven (11) weeks, and then bred to produce the F_2 litters."

a. Exposure: Rats were exposed to the test atmosphere for six hours per day, seven days per week for ten weeks prior to breeding, during

9

breeding and for the first 19 days of gestation. Females were not exposed from gestational day 20 until postnatal day 5. Beginning on postnatal day 5, exposure resumed; mothers were separated from their pups for the exposure period. Exposure of the females continued until sacrifice at weaning of the litters. Males were sacrificed following successful mating.

b. Mating: One male was caged with one female from the same test group until a vaginal plug or sperm in a vaginal smear were observed. If no evidence of successful mating was observed within 10 days, the male was removed and replace with a new male. Mating of brother-sister pairs was avoided.

Mated females were moved to solid bottom shoebox cages on day 19 of gestation.

c. Animal Assignment: F_0 Rats were assigned randomly, stratified by body weight to treatment groups.

4. Statistical Analysis: Per the report, "The unit of comparison was the male, the female, or the litter". "Results of the quantitative continuous variables (e.g., body weights, food consumption, organ weights, etc.) were intercompared for the three treatment groups and one control group by use of Levene's test for equal variances"; analysis of variance (ANOVA), and t-tests. When Levene's test indicated homogeneous variances and the ANOVA was significant, the pooled t-test was used for pairwise comparisons. When Levene's test indicated heterogeneous variances, all groups were compared by an ANOVA for unequal variances" "followed, when necessary, by the separate variance t-test for pairwise comparisons. The significance levels for the t-test comparisons were corrected by the Bonferroni method for all reproductive data."

"Nonparametric data were statistically evaluated using the Kruskal-Wallis test" "followed by the Mann-Whitney U test for pairwise comparisons" " when appropriate. Frequent data (such as the various indices) were compared using the Fisher's exact test". "For all statistical tests, the fiducial limit of 0.05 (two-tailed) was used as the criterion for statistical significance."

5. Compliance: Signed "Statement of No Data Confidentiality Claims", "Statement of Compliance" with 40 CFR Part 160 (GLP's) and Quality Assurance Unit audit statements were included.

II. RESULTS

A. Chamber Concentrations: During the first 82 exposure days of the study, the laboratory attempted to monitor the chamber concentration of PDCB by direct sampling through stainless steel tubing into gas tight syringes for introduction into the gas chromatograph for analysis. Concentrations obtained by this method for the first 40 days were only 68.0 - 80.0% of the nominal values. Analytical concentrations were further reduced during the next two sampling periods. The test laboratory took

10

this as an indication of a problem with the analytical or sampling technique used. Therefore, from day 83 of exposure through day 171, direct syringe sampling was substituted (no stainless steel tubing). Values obtained during this period were substantially higher than those previously recorded, confirming that a substantial sampling error existed for data collected from day 1 through day 82. For the remainder of the study, PDCB samples were collected by charcoal tube sampling.

The study authors suggest that the sampling problem arose because of the physical nature of the test material, which is a solid at room temperature. They suggest that the test material condensed on the interior surfaces of the tubing, resulting in lower apparent concentrations than were actually occurring in the chamber. When the tubing was eliminated from the sampling train, the analytical concentrations increased markedly, supporting the authors' contention.

The authors back-calculated to obtain what they describe as more accurate estimates of the test material concentrations from the first 171 days of exposure using the nominal concentrations and the ratios of analytical to nominal concentration from the charcoal tube sampling period. They state that the actual chamber concentrations for the entire 284 day exposure period were approximately 0.0, 66.3 ± 8.47, 211 ± 18.1 and 538 ± 50.5 ppm. They suggest that they were, at worst, always in excess of the nominal concentration, and therefore provide an adequate margin to ensure that the data present a conservative picture of the potential effects of exposure to the test material.

Based upon this report, no data exist which present an accurate picture of the exposure conditions which existed during this study. This includes the last third of the exposure period when charcoal tube samplers were used. Due to the high freezing point of the test material, it is impossible to determine how much of the test material condensed out on tubing or the coats of the rats. The exposure of the animals to PDCB may have been largely by the oral route.

CHAMBER ANALYSIS

Target Concentration (ppm)	0.0	50.0	150.0	450.0
Analytical Concentration (ppm)[a]	<MDL[b]	66.3 ± 8.47	211 ± 18.0	538 ± 50.5

a Grand mean of 284 daily means ± standard deviation.
b Less than the minimum detection limit of 1 ppm (all control chamber samples were <MDL except on study days 193, 194 and 218, when concentrations of 1, 3 and 1 ppm, respectively, were detected in the control chamber).

//

B. Parental Generations

1. Clinical Observations - Toxic clinical signs in F_0 male and female parents were only observed in rats exposed to 450 ppm atmospheres. These included increased incidences of tremors, unkempt appearance, urine stains, wetness of the fur, salivation and periocular, perinasal and perioral encrustations. High dose females also had increased incidences of wetness of the urogenital area.

Similarly, only high dose F_1 male and female parents showed marked increases in toxic clinical signs. The most notable of these included hypoactivity, ataxia, twitching, tremors, dehydration, unkempt appearance, lacrimation, periocular and perioral encrustation and perioral wetness. High dose females also exhibited increased urogenital area wetness.

MEASURED CHAMBER CONCENTRATION OF PDCB
(% of Nominal)

Time (days)	Nominal Concentration (ppm) 0	50	150	450
1 - 40	0	69.0	68.0	80.0
41 - 60	0	64.0	66.0	73.0
61 - 82	0	63.0	63.0	66.0
83 - 171	0	89.0	111.0	114.0
172 - 284	0	86.0	96.0	90.0

2. Mortality - Five adult animals died during the study and are listed below. None of the deaths appeared to be treatment related.

F_0 adult deaths:
Male #18817, 0.0 ppm, urinary tract inflammation and obstruction, day 83
Female #19013, 0.0 ppm, sacrificed moribund, day 50
Female #18988, 150.0 ppm, found dead, day 103
Female #18911, 450.0 ppm, found dead, day 97

F_1 adult deaths:
Male #35768, 50.0 ppm, sacrificed moribund, day 53

3. Body Weight - Premating body weights of female F_0 rats were unaffected by treatment with PDCB. High dose F_0 males had reduced body weights relative to the control by the end of week 1 of treatment and continuing through the remainder of the exposure period.

High dose F_1 female parents had significantly lower body weights than their control, low dose and intermediate dose counterparts at the initiation of the premating exposure period ($P \leq 0.01\%$). High dose females weighed only 85-90% as much as control females. Body weights for high

dose females remained significantly lower than the control through the entire premating exposure, but the relative difference remained constant. Therefore, this difference was an artifact of low birth weight. Similar results were observed in F_1 male parents. Although the high dose males were significantly lower in body weight than the controls at the initiation of the premating period and remained lower through mating, the degree of difference was relatively unchanged with the high dose weighing about 85% of the control at initiation and termination.

Gestational body weights of high dose F_0 females decreased due to treatment beginning at day 7; divergence from the control increased in magnitude until the end of gestation. The difference from control was not statistically significant (P<0.01%) until day 20. Gestational body weights of intermediate dose females showed similar decreases relative to the control but of lesser magnitude.

Gestational body weights of F_1 females were unaffected by treatment. At day 0 of gestation, high dose females had significantly lower body weights than their counterparts on other dosing regimes. This was an artifact of their lower birth weights and not a direct reflection of treatment. Throughout gestation, body weights of the high dose group remained significantly lower than the control but the relative difference remained unchanged at approximately 90%.

PARENTAL BODY WEIGHTS - MALES (g)

Time (Weeks)	Nominal Concentration (ppm) 0	50	150	450
F_0 Generation				
0	199.9	199.9	199.2	200.2
4	353.9	348.6	350.2	328.9**
8	474.2	466.2	474.2	428.9**
12	542.0	511.3	547.0	459.1**
16	592.3	596.1	603.1	536.9**
F_1 Generation				
0	226.9	218.7	230.6	192.9**
4	370.2	353.3	362.6	294.3**
8	463.4	443.6	462.0	381.9**
12	522.3	500.5	528.8	426.0**
16	565.6	538.7	569.7	481.8**

**Significantly different from the control (P≤0.01)

PARENTAL BODY WEIGHT GAINS - MALES (g)

Time (Weeks)	Nominal Concentration (ppm) 0	50	150	450
F_0 Generation				
0 - 4	153.9	148.7	151.0	128.6
4 - 8	120.3	117.6	123.9	100.0
8 - 12	67.8	45.1	72.8	30.3
12 - 16	48.9	84.8	56.0	77.8
0 - 16	392.4	396.2	404.2	336.7
F_1 Generation				
0 - 4	143.2	134.6	132.0	101.3
4 - 8	93.3	90.3	99.4	87.6
8 - 12	58.8	58.1	66.8	44.1
12 - 17	65.6	56.3	58.7	55.9
0 - 17	360.9	339.3	356.9	288.9

GESTATIONAL BODY WEIGHTS (g)

Time (Days)	Nominal Concentration (ppm) 0	50	150	450
F_0 Females				
0	287.84	281.21	277.33	277.28
7	319.02	312.93	310.96	304.30
13	350.59	341.55	339.90	330.85
20	422.11	410.00	402.09*	395.03**
F_1 Females				
0	306.39	291.99	308.48	270.79**
7	337.41	320.91	338.13	301.82**
13	356.47	342.74	359.94	321.40**
20	428.94	413.73	428.57	387.10**

*Significantly different from the control ($P \leq 0.05$)
**Significantly different from the control ($P \leq 0.01$)

Lactational body weights of F_0 females were not affected by treatment and did not differ from the control at any time. At delivery and until

the day 14 weighing, F_1 females had body weights that were statistically lower than the control. By day 14, the difference in body weight was still evident but not statistically significant. By day 28 of lactation, the body weights from all treatment groups were similar to the control, partially reflecting the simultaneous increase in body weight of the high dose group and the decrease in body weight of the other three groups.

GESTATIONAL BODY WEIGHT GAINS (g)

Time (Days)	Nominal Concentration (ppm) 0	50	150	450
F_0 Females				
0 - 7	31.19	31.72	33.64	27.02
7 - 13	31.56	28.62	28.94	26.54
13 - 20	71.52	68.45	62.19	64.18
0 - 20	134.27	128.79	124.76	117.75*
F_1 Females				
0 - 7	31.03	28.92	29.65	31.03
7 - 13	19.06	21.83	21.81	19.58
13 - 20	72.47	70.99	68.63	65.70
0 - 20	122.55	121.74	120.09	116.31

*Significantly different from the control ($P \leq 0.05$)

LACTATIONAL BODY WEIGHTS (g)

Time (Days)	Nominal Concentration (ppm) 0	50	150	450
F_1 Females				
0	333.41	322.94	331.96	298.09**
4	345.23	333.66	352.26	318.37*
7	346.59	338.24	353.01	319.15*
14	352.29	349.60	363.14	330.36
21	348.16	342.70	353.33	332.89
28	317.71	318.71	330.80	304.58

*Significantly different from the control ($P \leq 0.05$)
**Significantly different from the control ($P \leq 0.01$)

LACTATIONAL BODY WEIGHT GAINS (g)

Time (Days)	Nominal Concentration (ppm) 0	50	150	450
F_1 Females				
0 - 4	13.45	10.71	20.30	19.53
4 - 7	1.36	4.59	0.75	0.79
7 - 14	5.70	11.36	10.13	11.21
14 - 21	-4.13	-6.90	-9.81	5.20
21 - 28	-30.45	-24.00	-22.53*	-28.32
0 - 28	-14.07	-4.24	-1.16**	10.19**

*Significantly different from the control ($P \leq 0.05$)
**Significantly different from the control ($P \leq 0.01$)

4. Food Consumption - During the premating period for F_0 rats, decreased food consumption occurred in males and females only during the first week of exposure. For the duration of the premating period, no differences were observed.

During the premating period for F_1 parental rats, high dose males had significantly reduced food consumption from initiation of the premating exposure period through week 4 and again at week 9. During the remainder of the premating period, food consumption was reduced, but not to a statistically significant level. This observation is consistent with the lower body weights of this treatment group. Food consumption by F_1 parental females demonstrated no consistent treatment related effect during the premating period. Three isolated instances of statistically significant difference from the control in food consumption in high dose females were reported, however, they appeared to be numerical anomalies, unlinked in time or pattern of change.

C. Matings of the Parental Generations

1. Reproductive Performance - No clear treatment related effects on reproductive performance were reported. The latent time for mating for both generations was increased in the 450 ppm group, with the difference being more pronounced in the F_0 parents than in the F_1 parents. An increased percentage of matings occurred in these rats during the latter half of the mating period than in the controls. No other parameters reported were affected by treatment.

2. Neonatal Growth and Development - The total number of pups born per litter was not affected by treatment in either F_1 or F_2 litters; however, live births were slightly reduced in F_2 pups ($P \leq 0.01\%$). Four day survival was reduced in high dose F_1 and F_2 pups, but not affected in low or intermediate dose pups. At day 4, litters were culled to yield as

NUMBER (%) OF SPERM- OR PLUG-POSITIVE FEMALES

Time (Days)	Nominal Concentration (ppm) 0	50	150	450
F_0 Females				
First 10 Days	24 (88.9)	27 (96.4)	28 (100.0)	21 (75.0)
Last 11 Days	3 (11.1)	1 (3.6)	0 (0.0)	7 (25.0)
F_1 Females				
First 10 Days	27 (100.0)	26 (92.9)	25 (89.3)	23 (82.1)
Last 11 Days	0 (0.0)	2 (7.1)	3 (10.7)	5 (17.9)

nearly as possible four males and four females. Additional pups were sacrificed. No treatment related effects on survival were reported on pups after day 4.

High dose pups from both generations had significantly reduced body weights at birth ($P \leq 0.01\%$) relative to the control. Pups from other treatments were similar to the control. Differences in the body weights of high dose pups persisted through the 28 day lactation period.

SUMMARY OF LITTER SIZE

Lactational Day		Nominal Concentration (ppm) 0	50	150	450
F_1 Pups					
0	Total Born/Litter	13.9	14.1	12.0	12.5
	Total Live/Litter	13.0	14.0	11.6	11.6
4	Total Live/Litter	12.9	13.7	11.2	10.5*
F_2 Pups					
0	Total Born/Litter	13.5	12.8	13.7	11.4
	Total Live/Litter	14.0	12.5	13.6	10.7**
4	Total Live/Litter	13.9	12.4	13.5	10.7**

*Significantly different from the control ($P \leq 0.05$)
**Significantly different from the control ($P \leq 0.01$)

17

LITTER VIABILITY

Lactational Day		Nominal Concentration (ppm) 0	50	150	450
F_1 Pups					
0	Total Born	313	324	323	276
	Total Born Alive	313	323	313	256
	No. StillBorn	34	1**	10**	20
4	No. Alive	296	315	292	209
	No. Dead	17	8	21	47**
F_2 Pups					
0	Total Born	310	255	329	239
	Total Born Alive	308	249	326	225
	No. Stillborn	2	6	3	14**
4	No. Alive	305	248	323	171
	No. Dead	3	1	3	54**

**Significantly different from the control ($P \leq 0.01$)

D. Necropsy Findings: No treatment related observations were reported with the exception of organ weight effects.

The absolute liver weights of F_0 male rats were significantly increased in mid and high dose groups. Liver weights expressed as percent of body weight were increased in a dose related fashion in all treated groups; expressed as percent of brain weight, liver weights increased in the mid and high dose groups. Absolute kidney weights, kidney weights expressed as percent of body weight and expressed as percent of brain weight were significantly increased in a dose related fashion in all treated groups. Brain and testes weights were not affected by the test material.

Organ weight effects in F_1 males were less pronounced than in F_0 males. Absolute liver weights were increased only in the high dose group. When expressed as percent of body weight and as percent of brain weight, liver weights increased in the mid and high dose groups although the increase as percent of brain weight was not statistically significant in the mid dose males. Absolute kidney weights increased significantly in mid and high dose F_1 males. When expressed as percent of body weight and percent of brain weight, kidney weights increased in a dose related fashion in all treated groups although the increase as percent of brain weight in the low dose group was not statistically significant. Brain and testes weights expressed as percent of body weight were significantly increased ($P<0.01\%$) in high dose F_1 males only.

MEAN PUP BODY WEIGHTS - ENTIRE LITTER (g)

Lactational Day	Nominal Concentration (ppm) 0	50	150	450
F_1 Pups				
0	6.14	5.98	6.08	5.37**
4 (Pre Cull)	10.67	10.15	10.66	8.84**
4 (Post Cull)	10.72	10.25	10.78	8.90**
7	15.34	14.91	15.35	12.02**
14	27.99	27.12	27.56	22.04**
21	46.57	44.79	45.96	36.47**
28	83.37	79.91	82.21	67.81**
F_2 Pups				
0	6.23	6.32	6.19	5.43**
4 (Pre Cull)	10.52	10.68	10.47	8.11**
4 (Post Cull)	10.49	10.67	10.52	8.22**
7	15.19	14.96	15.46	11.60**
14	28.25	27.78	27.72	21.88**
21	45.26	44.27	45.36	37.86**
28	83.22	81.84	83.79	69.94**

**Significantly different from the control ($P \leq 0.01$)

For F_0 female rats, absolute liver weights, liver weights expressed as percent of body weight and expressed as percent of brain weight were increased in the 150 ppm and 450 ppm treatment groups. Increases in absolute liver weights and liver weights expressed as percent of brain weight in the low dose females were not statistically significant. Absolute and relative kidney weights of high dose females only were increased due to treatment although the increase was not statistically significant when kidney weights were expressed as percent of brain weight. Ovary weights when expressed as percent of brain weight were increased although not significantly.

No effects on kidney weight were seen in any treated group of F_1 female rats. High dose females had significantly increased absolute and relative body weights; mid dose females had increased absolute liver weights and liver weights as percent of brain weight although the differences presented were not statistically significant.

2. Clinical Pathology Findings - No treatment related gross pathological findings were reported.

SUMMARY OF ORGAN WEIGHT DATA

	Nominal Concentration (ppm) 0	50	150	450
F_0 Adult Males				
LIVER				
Organ weight (g)	20.545	21.703	23.799**	28.269**
% of body wt.	3.465	3.631*	3.945**	5.271**
% of brain wt.	936.901	1013.791	1107.892**	1284.090**
KIDNEY				
Organ weight (g)	3.984	4.475**	4.854**	5.192**
% of body wt.	0.675	0.752**	0.806**	0.970**
% of brain wt.	181.709	208.848**	225.913**	235.934**
F_1 Adult Males				
LIVER				
Organ weight (g)	21.191	20.036	22.557	24.829**
% of body wt.	3.593	3.588	3.835*	5.163**
% of brain wt.	976.517	942.114	1059.074	1159.987**
KIDNEY				
Organ weight (g)	3.908	4.142	4.566**	4.753**
% of body wt.	0.667	0.745**	0.777**	0.987**
% of brain wt.	180.060	195.057	214.002**	222.141**
F_0 Adult Females				
LIVER				
Organ weight (g)	13.068	13.401	14.271	17.106**
% of body wt.	4.096	4.258	4.480*	5.367**
% of brain wt.	669.350	669.403	711.505	979.473**
KIDNEY				
Organ weight (g)	2.354	2.358	2.430	2.649*
% of body wt.	0.745	0.752	0.764	0.830**
% of brain wt.	120.896	117.992	121.147	162.990
F_1 Adult Females				
LIVER				
Organ weight (g)	13.398	13.671	14.327	15.537**
% of body wt.	4.153	4.274	4.319	4.969**
% of brain wt.	678.786	684.446	724.041	781.016**
KIDNEY				
Organ weight (g)	2.366	2.384	2.426	2.384
% of body wt.	0.734	0.747	0.732	0.764
% of brain wt.	119.815	119.290	122.613	120.056

*Significantly different from the control ($P \leq 0.05$)
**Significantly different from the control ($P \leq 0.01$)

20

3. Microscopic Pathology Findings - Histopathological effects of the test material in adult female rats were limited to significantly increased hepatocellular hypertrophy in the high dose group. The response in F_1 females (14/28, $P \leq 0.01\%$) was more pronounced than in F_0 females (7/27, $P \leq 0.05\%$).

Similar liver effects were seen in high dose, adult males from the F_0 and F_1 generations. Hepatocellular hypertrophy was significantly increased in both groups ($P \leq 0.01\%$), and hepatocellular swelling (F_1) and hepatocellular cytoplasmic vacuolation (F_0) were increased although not significantly. In F_0 males only, mineralized bodies and prostatitis were significantly ($P<0.01\%$) decreased relative to the control. The biological significance of this observation is unclear; as the effect was not repeated in the F_1 males, it may be a numerical anomaly. The kidneys of adult, male F_0 and F_1 rats reflected the most signifcant adverse response to exposure to the test material. All treated male rats exhibited hyaline droplet nephrosis. Although this lesion also occurred in 11/27 and 10/28 control rats, the severity was increased in the treated animals in addition to the frequency. Control rats had mild hyaline droplet nephrosis in contrast to marked effects in treated animals at all treatment levels. A number of other kidney lesions were reported in males exposed to the test material. These effects were increased in both generations but appeared to be more pronounced in the F_0 males than in the F_1 males.

No treatment related histopathological effects were reported in F_2 weanlings of either sex.

No treatment related histopathological effects were reported for the reproductive organs of any group in this study.

III. DISCUSSION: Exposure to the test material was found to result in clinical signs of toxicity only in high dose adults. However, at necropsy, organ weight changes and microscopic pathology indicated toxic effects at the mid and low dose. In particular, abnormal pathology of the kidneys of adult males was observed. This lesion did not occur in females. The authors cited a literature reference which reported similar kidney lesions from exposure of male Fischer 344 rats to p-dichlorobenzene subchronically at doses of 150 - 600 mg/kg/day (Bomhard, E., Luckhaus, G., Voight, W.-H., and Loesner, E. 1988. Induction of light hydrocarbon nephropathy by p-dichlorobenzene. Arch. Toxicol. 61:433-439). In an article by Alden (Alden, C. L. 1986. A review of unique male rat hydrocarbon nephropathy. Toxicol. Pathol. 14:109-111), hyaline droplet nephrosis was postulated to develop due to enhanced absorption of $\alpha_{2\mu}$-globulin by the proximal tubule. An alternate hypothesis offered was decreased lysosomal catabolism of $\alpha_{2\mu}$-globulin in tubular epithelial cells due to binding of the test material (Charbonneau, M. and Swenberg, J. A. 1988. Studies on the biochemical mechanism of $\alpha_{2\mu}$-globulin nephropathy in rats. CIIT Activities 8(6):1-5; Short, B. and Swenberg, J. A.

MICROSCOPIC PATHOLOGY IN ADULT RATS
TERMINAL SACRIFICE

Observation	Nominal Concentration (ppm) 0	50	150	450
F_0 Males				
LIVER (examined 27, 28, 28, 28)				
Hepatocellular cytoplasmic vacuolation	0	0	0	3
Hepatocellular hypertrophy	0	1	1	27**
PROSTATE (examined 27, 0, 0, 28)				
Mineralized bodies	11	0	0	2**
Prostatitis	9	0	0	1**
KIDNEYS (examined 27, 28, 28, 28)				
Tubular degeneration	0	0	1	3
Hyaline droplet nephrosis	11	27**	28**	28**
Tubular proteinosis	1	12**	11**	22**
Granular cast formation	0	10**	15**	22**
Nephritis, interstitial	2	9*	14**	21**
Fibrosis, interstitial	0	6*	8**	5
Tubular cell hyperplasia/ hypertrophy	0	4	5	16**
F_0 Females				
LIVER (examined 27, 28, 27, 27)				
Hepatocellular hypertrophy	0	0	0	7*
F_1 Males				
LIVER (examined 28, 27, 28, 28)				
Hepatocellular swelling	0	0	0	5
Hepatocellular hypertrophy	0	0	0	21**
KIDNEYS (examined 28, 27, 28, 28)				
Hyaline droplet nephrosis	10	27**	28**	28**
Tubular proteinosis	1	2	8*	15**
Granular cast formation	0	2	16**	16**
Nephritis, interstitial	4	9	14**	25**
Fibrosis, interstitial	1	2	6	5
Tubular cell hyperplasia/ hyperplasia	0	1	4	7*
F_1 Females				
LIVER (examined 28, 28, 28, 28)				
Hepatocellular hypertrophy	0	0	0	14**

*Significantly different from the control ($P \leq 0.05$)
**Significantly different from the control ($P \leq 0.01$)

22

1988. Pathologic investigations of the mechanism of unleaded gasoline-induced renal tumors in rats. CIIT Activities 8(7):1-6). Because $\alpha_{2\mu}$-globulin occurs only in post-pubertal male rats, the data in this report are consistent with these hypotheses.

Gestational body weights were significantly decreased in F_0 and F_1 females on the 450 ppm diet. These females produced litters with reduced numbers of live pups, reduced body weight of pups and decreased survival of pups by day 4 of lactation. No developmental effects were observed in the absence of maternal toxicity.

This study contains two major design flaws which limit its usefulness in evaluating the reproductive hazard presented by inhalation of the test material. First, due to the poor design of the atmosphere sampling system, the concentrations of material to which the rats were exposed cannot accurately be determined. A second major difficulty in evaluating the inhalation effects of this compound arises from the physical characteristics of the compound. This material has a melting point of 52°C and condenses upon cooling onto any surrounding surface. Although sufficient material was taken in by the rats to elicit definite toxic effects, the material may largely have been ingested rather than inhaled.

IV. CONCLUSIONS: Paradichlorobenzene caused hyaline droplet nephrosis in male rats at concentrations ≥ 50 ppm in the test atmosphere. Exposure of pregnant female rats to atmospheres containing 450 ppm PDCB resulted in litters with reduced numbers of live pups, reduced pup weights and decreased pup survival at day 4 of lactation.

NOEL for Reproductive Toxicity = 150 ppm based on litters with reduced numbers of live pups, pup weights and 4-day survival
LOEL for Reproductive Toxicity = 450 ppm
NOEL for Systemic Effects < 50 ppm
LOEL for Systemic Effects = 50 ppm based on hyaline droplet nephrosis in male rats

V. CLASSIFICATION: **Core - Supplementary**

Deficiencies:
1) The sampling techniques for measuring chamber concentrations were inappropriate and changed during the 284 day study period. Therefore, the concentrations of paradichlorobenzene in the chambers were not adequately determined.
2) None of the concentrations used in this study constituted a systemic NOEL.

This study does not satisfy the guideline requirements (83-4) for a "Two Generation Reproduction Study in Rats".

23