AIM

Association for Information and Image Management

1100 Wayne Avenue, Suite 1100
Silver Spring, Maryland 20910
301/587-8202

Centimeter

Inches

MANUFACTURED TO AIM STANDARDS
BY APPLIED IMAGE, INC.

1 of 3

NOTICE

This report was supported in part, or whole, by funds from the Comprehensive Environmental Response, Compensation, and Liability Act trust fund by interagency agreement with the Agency for Toxic Substances and Disease Registry, United States Public Health Service.

RECEIVED

MAY 31 1994

OSTI

**Inhalation Developmental
Toxicology Studies:
Developmental Toxicity
of Chloroprene Vapors in
New Zealand White Rabbits**

T. J. Mast
J. J. Evanoff
R. B. Westerberg

R. L. Rommereim
R. J. Weigel

April 1994

Prepared for the National Institute
of Environmental Health Sciences,
National Toxicology Program
under a Related Services Agreement
with the U.S. Department of Energy
Contract DE-AC06-76RLO 1830

Pacific Northwest Laboratory
Operated for the U.S. Department of Energy
by Battelle Memorial Institute

PNL-7458

DISTRIBUTION OF THIS DOCUMENT IS UNLIMITED

DISCLAIMER

This report was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency thereof, nor Battelle Memorial Institute, nor any of their employees, makes **any warranty, expressed or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights.** Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof, or Battelle Memorial Institute. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

PACIFIC NORTHWEST LABORATORY
operated by
BATTELLE MEMORIAL INSTITUTE
for the
UNITED STATES DEPARTMENT OF ENERGY
under Contract DE-AC06-76RLO 1830

Printed in the United States of America

Available to DOE and DOE contractors from the
Office of Scientific and Technical Information, P.O. Box 62, Oak Ridge, TN 37831;
prices available from (615) 576-8401. FTS 626-8401.

Available to the public from the National Technical Information Service,
U.S. Department of Commerce, 5285 Port Royal Rd., Springfield, VA 22161.

The contents of this report were printed on recycled paper

"This report was supported in part, or whole, by funds from the Comprehensive Environmental Response, Compensation, and Liability Act trust fund by interagency agreement with the Agency for Toxic Substances and Disease Registry, United States Public Health Service."

INHALATION DEVELOPMENTAL TOXICOLOGY STUDIES: DEVELOPMENTAL
TOXICITY OF CHLOROPRENE VAPORS IN NEW ZEALAND WHITE RABBITS

Final Report
No. NIH-Y01-ES-70153

T.J. Mast, Principal Investigator
J.J. Evanoff
R.B. Westerberg

R.L. Rommerein
R.J. Weigel

April 1994

Prepared for the National Institute of
Environmental Health Sciences, National
Toxicology Program under a Related
Services Agreement with the U.S.
Department of Energy under Contract
DE-AC06-76RLO 1830.

Pacific Northwest Laboratory
Richland, WA 99352

MASTER

DISTRIBUTION OF THIS DOCUMENT IS UNLIMITED

for

PEER REVIEW STATEMENT

This final technical report has been peer reviewed by two scientists who were members of the Reproductive and Developmental Toxicology Program Review Subcommittee for the National Toxicology Program Board of Scientific Counselors. Reviewers serve as independent scientists, not as representatives of any institution, company, or government agency. In this capacity, these reviews determine if the design and conduct of the NTP studies were appropriate, ensure that the final report presents the experimental results, and that conclusions are consistent with the data.

Reviewers of this developmental toxicity study of chloroprene were:

Dr. George Szczech
Burroughs-Wellcome
Research Triangle Park, NC

Dr. Robert Kavlock
U.S. EPA
Research Triangle Park, NC

These reviewers in general agreed that the study was properly designed except one reviewer noted that higher concentrations of inhaled chloroprene would have made this a more rigorous study in rabbit. Both reviewers requested more background information and discussion on the occurrence and significance of variations in the conformation of lung lobes of fetuses. However, no background information on the lobulation of rabbit lungs was found in a search of the literature. This final report takes reviewer's comments into account and incorporates their suggestions on minor points and the format of the report.

B.A. Schwetz, D.V.M., Ph.D.
Project Officer

SUMMARY

Chloroprene, 2-chloro-1,3-butadiene, is a colorless liquid with a pungent ethereal odor that is primarily used as an intermediate in the manufacture of neoprene rubber, and has been used as such since about 1930. This study addressed the potential for chloroprene to cause developmental toxicity in New Zealand white rabbits following gestational exposure to 0, 10, 40, or 175 ppm chloroprene vapors, 6 h/dy, 7 dy/wk. Each treatment group consisted of ~15 artificially inseminated females exposed on 6 through 28 days of gestation (dg). Body weights were obtained throughout the study period, and uterine and fetal body weights were obtained at sacrifice on 29 dg. Implants were enumerated and their status recorded and live fetuses were examined for gross, visceral, skeletal, and soft-tissue craniofacial defects.

There were no overt signs of maternal toxicity and the change in maternal body weight over the course of the study was not affected. Exposure of pregnant rabbits to chloroprene vapors on 6-28 dg had no effect on the number of implantations, the mean percent of live pups per litter, or on the incidence of resorptions per litter. Fetal body, kidney, and liver weights (as means of litter means) were not affected by gestational exposure to chloroprene, nor was the fetal sex ratio affected. The incidence of fetal malformations was not increased by exposure to chloroprene. There were no significant alterations in the incidence of total fetal variations or reduced ossifications among exposed groups. Results of this study indicate that gestational exposure of New Zealand white rabbits to 10, 40, or 175 ppm chloroprene did not result in observable toxicity to either the dam or the offspring. The highest exposure group, 175 ppm, was the no observable adverse effect level (NOAEL) for rabbits in the study with respect to both maternal and developmental toxicity.

ACKNOWLEDGMENTS

<u>Responsibility</u>	<u>Name</u>
Principal Investigator	T.J. Mast
Exposure System	R.J. Weigel
	E.J. Rossignol
Monitoring/Analytical Chemistry	R.B. Westerberg
	K.H. Stoney
Animal Resources Center	M.G. Brown
	L.G. Gorham
	S.E. Rowe
	A.E. Jarrell
Developmental Toxicology	T.J. Mast
	J.J. Evanoff
	R.L. Rommereim
Health and Safety	M.L. Clark

This work was supported by the National Institute of Environmental Health Sciences, National Toxicology Program under a Related Services Agreement with the U.S. Department of Energy under Contract DE-AC06-76RLO 1830 at the Pacific Northwest Laboratory operated for the U.S. Department of Energy by Battelle Memorial Institute.

The excellent technical support in the Developmental Toxicology Laboratory of P.J. Boyd, S. Martell and R. Ballou is appreciated.

SIGNATURES

Terry J. Mast

Terry J. Mast, PhD, DABT
Principal Investigator and
Developmental Toxicology Task Leader

5/12/94

Date

R. Bruce Westerberg

R. Bruce Westerberg, PhD
Chemistry Task Leader

5/17/94

Date

Richard J. Weigel

Richard J. Weigel, PhD
Exposure Task Leader

5/17/94

Date

Stephen E. Rowe

Stephen E. Rowe, DVM, ACLAM
Laboratory Animal Medicine Veterinarian

5/17/94

Date

TABLE OF CONTENTS

INTRODUCTION	1
HUMAN DATA AND EXPOSURE	1
DEVELOPMENTAL TOXICITY	3
MATERIALS AND METHODS	7
ANALYTICAL CHEMISTRY	7
Bulk Chemical Analysis	7
Exposure Chamber Monitoring	7
Degradation and Stability Studies	8
GENERATION AND EXPOSURE METHODS	9
Exposure Chambers	9
Exposure Suite System Description	10
Generation and Delivery System	11
Characterization of Test Article Concentration in Chambers	12
HEALTH AND SAFETY	14
ANIMAL HUSBANDRY	15
DEVELOPMENTAL TOXICITY STUDY DESIGN	18
DATA ANALYSIS	20
DATA STORAGE	21
RESULTS	23
DISCUSSION	25
REFERENCES	27

LIST OF FIGURES

Figure 1	Inhalation Exposure Chamber Designed at PNL	29
Figure 2	Chloroprene Developmental Toxicity Study Exposure Suite ..	30
Figure 3	Chloroprene Vapor Generation and Distribution System	31
Figure 4	Buildup and Decay Curves of Concentration in Animal Chambers	32
Figure 5	Animal Exposure Chamber Schematic Showing Approximate Sampling Location	33
Figure 6	Post-Exposure Monitoring Results of the 175 ppm A Chloroprene Chamber	35
Figure 7	Chamber Relative Humidity Control System	36

LIST OF TABLES

Table 1	Summary of T ₉₀ and T ₁₀ Data	37
Table 2	Summary of Chamber Uniformity Data	38
Table 3	Summation of Exposure Concentration Data	39
Table 4	Summation of Chamber Environmental Data	40
Table 5	Study Disposition	41
Table 6	Summary of Gross Observations at Time of Maternal Sacrifice	42
Table 7	Mean Body, Uterine and Extra-gestational Gain and Organ Weights of Pregnant Developmental Toxicology Rabbits	43
Table 8	Reproductive Measures in Rabbits	44
Table 9	Fetal Organ and Body Weights, Fetal Organ to Body Weight Ratio, and Fetal Sex Ratio in Rabbits	45
Table 10	Malformations Observed in Live Rabbit Fetuses	46
Table 11	Variations and Reduced Ossifications Observed in Live Rabbit Fetuses	47
Table 12	Mean Percent of Live Rabbit Fetuses Affected per Litter ..	48

INTRODUCTION

Chloroprene, 2-chloro-1,3-butadiene, is a colorless liquid with a pungent ethereal odor. It is inherently unstable in the presence of oxygen at room temperature and reacts readily to form peroxides and radicals which lead to rapid polymerization. In order to prevent these reactions from taking place two inhibitors are commonly added to bulk chloroprene, *t*-butylcatechol (TBC) and phenothiazine. Chloroprene is produced by reacting butadiene with chlorine to form a mixture of dichlorobutene isomers (IARC 1979), from which 3,4-dichloro-1-butene is isolated and reacted with caustic soda to form chloroprene.

Chloroprene is used primarily as an intermediate in the manufacture of neoprene rubber, and has been used as such since about 1930. Circa 1976, United States production of chloroprene was approximately 164 million kg, about 100 million kg in Western Europe, and 80 million kg in Japan (IARC 1979). Chloroprene is also produced in the USSR and in China; however, no production figures are available for these countries.

The inhalation and dermal routes are relevant to occupational exposure to chloroprene. The current OSHA standard for chloroprene in air averaged over an 8-h work day is 25 ppm (80 mg/m³) with an accompanying notation to avoid skin contact, however the ACGIH recommended TLV-TWA is only 10 ppm (35 mg/m³) also with a recommendation to avoid dermal exposure. Infante et al. (1977) estimated that approximately 2500-3000 United States workers are exposed to chloroprene. They reported airborne chloroprene concentrations ranging from 6 to 1420 ppm in various areas of a U.S. chloroprene polymerization plant with the greatest concentrations in the make-up and reactor areas.

HUMAN DATA AND EXPOSURE

Acute exposure to chloroprene vapors results in irritation to the skin, eyes, and mucous membranes of the respiratory tract, and dermal exposure has

been reported to cause loss of hair (Sittig, 1985). Exposure to higher concentrations causes excessive nervousness and irritability followed by central nervous system effects and respiratory depression (IARC 1979).

Chronic exposure to chloroprene, aside from possible carcinogenicity and reproductive toxicity, may cause CNS depression and damage to the lungs, kidneys, and liver in both humans and experimental animals (Plunkett 1987 and others). Damage to the myocardium in humans has also been reported (Sittig 1985). Evaluation of biochemical and hematological parameters in workers did not indicate or suggest that chloroprene caused alterations of medical significance (Gooch and Hawn 1981).

A large number of studies on the effects of occupational exposure to chloroprene have been performed in Russia and are cited and summarized in an IARC monograph (1979). Reported effects of chronic exposure to chloroprene are numerous and varied and thus, may represent the effect of exposure to a mixture of compounds. Some of the more specific effects include loss of hair, hepatomegaly, abnormal liver function, dystrophy of the myocardium, circulatory changes, anemia, dysfunction of both the central and peripheral nervous system, and periodontal disorders. One Russian study reported low immunological reactivity in more than 200 chloroprene-exposed workers who had been immunized with typhoid vaccine, and several studies reported an increased number of chromosome aberrations in cultured peripheral lymphocytes as well as functional disturbances in male reproductive function.

Metabolism:

The metabolism of chloroprene has not been studied in depth, but the formation of a reactive intermediate capable of alkylation *in vivo* was inferred by Bartsch et al (1979) after demonstrating the formation of adducts of 4-(4-nitrobenzyl)pyridine and chloroprene by mouse liver microsomes. These reactive intermediates may be formed by the epoxidation of one or both of the double bonds in chloroprene. Conjugation of chloroprene or a metabolite with glutathione has been shown to occur rapidly in Wistar rats (Summer and Greim 1980).

DEVELOPMENTAL TOXICITY

Several studies have been conducted in order to assess the potential for β -chloroprene to cause embryonic or developmental toxicity in laboratory animals. Salnikova (1968) investigated the embryotoxic effects of inhalation exposure to chloroprene and ammonia combined; however, since a group exposed to chloroprene alone was not included it is hard to interpret their results. In a later study, Salnikova and Fomenko (1973) exposed pregnant rats to 0.016, 0.36, 0.17, 0.83, and 1.11 ppm chloroprene vapor. The length and duration of the exposures was not specified. An increased incidence of embryonic deaths was reported in the 0.83 and 1.11 ppm groups, and fetuses in the 1.11 ppm group had a significantly lower birth weight than those in the control group. Fetuses in the 1.11 ppm group were also reported to have vascular permeability disturbances as well as decreases in the lengths of the tibia and the fibula. The methodology used for the generation of the chloroprene test atmospheres and the techniques used for monitoring the purity of the atmospheres (if any) were not specified.

Culik et al. (1978) exposed CD-1 (Sprague-Dawley) rats to 0, 1, 10, or 25 ppm chloroprene, 4 h/day, on 3 through 20 days of gestation (dg) to assess developmental toxicity. Another group was exposed to the same concentration on 1 through 12 dg in order to assess the embryotoxic potential of chloroprene. Care was taken during this study to ensure that the animals were exposed to chloroprene which was freshly distilled and relatively free of the toxic dimer. Dams exposed on 3-20 dg were killed on 21 dg and the fetuses examined for signs of developmental toxicity. Dams exposed on 1-12 dg were killed on 17 dg and the uterine contents evaluated for signs of embryotoxicity.

No clinical signs of toxicity were observed in the dams exposed to chloroprene, and there was no affect on weight gain with respect to the control animals. There was no effect on litter size, on the number of implantations, or the number of resorptions in exposed pregnant rats in either the teratology or the embryotoxicity study. Although fetal body weight and crown-rump length were greater for the 10- and 25-ppm groups than for the controls these effects were not thought to be treatment-related.

The Joint Industry Committee on Chloroprene contracted an inhalation developmental toxicity study on β -chloroprene in rats (Koeter and Appelman 1980). In this study pregnant rats were exposed to 0, 10, 25, 75, or 175 ppm chloroprene 6 hours/day, on 4-16 dg. They reported a diminished maternal weight gain during the exposure period for the 25, 75, and 175 ppm group, and during the post-exposure period for the 75 and 175 ppm groups. Post-implantation loss was significantly greater than controls in the 10, 75, and 175 ppm groups, but not in the 25 ppm group. There were fewer live fetuses in the two highest exposure groups than in the controls; however, since there was no apparent dose-response relationship the authors did not attribute this effect to chloroprene exposure.

There was a significant reduction in fetal weight in the 75 and 175 ppm groups, but no abnormalities attributable to treatment were found in the fetuses from any of the exposed groups. There was a statistically significant increase in the incidence of unossified forelimb phalanges and metatarsals in the 25, 75 and 175 ppm groups, and the incidence of unossified cervical vertebral bodies was increased in the 10, 75, and 175 ppm groups. The authors assert there is no clear dose-response relationship and apparently do not view these effects as being toxicologically significant since they are not reported in the study summary.

In contrast to the Russian studies (Salnikova and Fomenko 1973), chloroprene did not appear to cause developmental or reproductive toxicity at exposure concentrations as high as 25 ppm under the conditions of Culik et al. (1978). However, since a maternally toxic dose was not achieved, and 25 ppm chloroprene is the OSHA TLV, a margin of safety is not established by this study. Although the data from the Russian studies indicate that chloroprene may be considerably more toxic than implied by Culik et al. (1978), this disparity may be due to the formation of dimers and other toxic reaction products in the exposure atmospheres used by the Russians. Test material generation and monitoring methods were not described by the Russian authors although it is well known that chloroprene rapidly degrades in air to form more toxic products (von Oettingen et al. 1936, and others). In the study performed by Koeter and Appelman (1980), higher exposure concentrations were used and care was taken to ensure that the chloroprene used for exposures was

freshly prepared and properly stored. Test atmospheres were not monitored for dimer concentrations in this study.

The study in rabbits described in this report was conducted in an attempt to clarify the ambiguities regarding the potential for chloroprene to cause embryonic and developmental toxicity in laboratory models. To date only one laboratory specie, the rat, has been well-tested for developmental toxicity. In order to facilitate an interspecies comparison of the data, exposure concentrations similar to those of Koeter and Appelman (1980) will be used in this study. The lowest concentration, 10 ppm, is the current TLV for chloroprene, while the highest concentration, 175 ppm, would be exceeded only in accidental situations. This study was designed not so much to evaluate the potential developmental toxicity in the rabbit, but essentially to compare the sensitivity to chloroprene between two different animal models, rabbits and rats, at or above potential occupational exposure levels to humans.

MATERIALS AND METHODS

Four groups of New Zealand white rabbits were exposed to 0, 10, 40 or 175 ppm chloroprene vapors, 6 h/day, 7 days/week, on 6 through 28 days of gestation (dg). Developmental evaluations were conducted on pregnant rabbits sacrificed on 29 dg.

ANALYTICAL CHEMISTRY

Chloroprene (Denka Chemical Corporation, Houston, Texas) was received by Pacific Northwest Laboratory (PNL) on February 14, 1990 (BNW Lot No. 53436-20). The test material was inhibited with $\approx 0.1\%$ each of phenothiazine and t-butylcatechol (4-TBC) at Denka and shipped at ambient temperature. The daily requirement of chloroprene for exposure generation was ≈ 0.6 kg, and ≈ 15 kg of test material was consumed during the study. Prior to use, test material was maintained at approximately -20°C in the LSL-II storage facility. All transfers of chloroprene were performed under nitrogen to prevent the introduction of air into the bulk chemical.

Bulk Chemical Analysis

Bulk chemical analyses of the test material were performed using infrared spectroscopy to confirm test material identity. Purity and dimer determinations were conducted by gas chromatography (GC). Test material purity was acceptable throughout the study. Peroxide determinations were made by potentiometric titrations of samples with standardized sodium thiosulfate. Chloroprene was analyzed for peroxide content within 6 weeks prior to use as test material and contained ≤ 1 meq/kg at each analysis. A specification for a maximum peroxide content of 1 meq/kg was applied throughout to ensure the safety of the generation process which involved evaporating chloroprene at a temperature of $\approx 150^{\circ}\text{C}$. (See Appendix A for details.)

Exposure Chamber Monitoring

A Hewlett-Packard (HP) 5890 GC system (1/8" o.d. x 1.0' nickel column packed with 1% SP-1000 on 60/80 mesh Carbopack B; 120°C) was used to monitor chambers during animal exposures (Figure 1). This instrument was equipped

with a 12-port stream select valve and measured chloroprene in the exposure chambers, the control chamber, the exposure room, the on-line standard, and the filtered air blank. An on-line, certified standard, 80 ppm chloroprene in nitrogen (MG Industries Scientific Gases, Los Angeles, CA), was used to check instrument drift throughout the exposure day. In order to confirm values obtained from the on-line monitor, bubbler samples were collected and analyzed with an HP 5830 or HP 5840 GC equipped with a glass column packed with 10% Carbowax 20 M on 80/100 Chromasorb.

The precision of the on-line GC was estimated from measurements of the on-line chloroprene standard; a 0.5% coefficient of variation was observed. Linearity of the on-line GC was assured by calibrating it against a gravimetrically calibrated GC. The minimum detectable limit of the GC was determined from precision measurements performed by monitoring a chamber with a low chloroprene concentration and was estimated to be 0.06 ppm chloroprene. (See comments in the "Test Chemical Concentration Monitoring" section in Appendix A).

Degradation and Stability Studies

The stability of chloroprene and the extent to which less volatile impurities and peroxides concentrated in the generator flask over the 6-hour exposure period were evaluated during prestart testing. A chloroprene sample was removed from the generation flask immediately after filling at the beginning of the day and an additional sample was collected at the end of the exposure period.

Chloroprene was determined to be 98.5% pure by area percent in the generation flask immediately after filling, and 90.6% at the end of generation on the same day. The most concentrated dimer isomer was present at ~2% while the total dimer content was about 5-6%. The peroxide content in the sample collected immediately after filling was 0.05 meq/kg, while the peroxide content at the end of generation on the same day was 0.14 meq/kg. These analyses indicated that the purity of test material in the generation flask at the end of the day was substantially lower than the purity at the beginning of

the day due to formation and concentration of dimer isomers and peroxides in the generation flask.

The purity of chloroprene vapor directed to the exposure chambers was monitored every exposure day using a separate HP 5890 GC (on-line purity GC). The on-line purity GC was used to ensure that volatile compounds were vented to waste before initiation of animal exposures. Once exposures were initiated, the GC parameters were changed to allow both volatile and less volatile impurities (including dimers) to be determined. The on-line purity GC showed an average percent purity of 99.34% for the study.

Chamber atmospheres and the vapor distribution line were tested for the presence of trace amounts of contaminants or degradation products by gas chromatography. Assignment of dimer and inhibitor peaks was verified by gas chromatography/mass spectrometry. Trace amounts of chlorobutene and 1,3-chlorobutadiene (<0.8% by area) were found in samples collected from the chambers and the distribution line. The inhibitors, 4-TBC and phenothiazine were present in the distribution line at ≈ 3 ppm. Dimers were not detected in the distribution or chamber samples by either the analysis of charcoal tube samples or the on-line purity method. The detection limits for dimer were estimated to be $\approx 0.02\%$ by the charcoal tube method, and $\approx 0.04\%$ by the on-line method (as percent of the total peak area of the most abundant dimer isomer). Test chemical stability and purity were considered acceptable for the study.

GENERATION AND EXPOSURE METHODS

Exposure Chambers

The animals were exposed and maintained in inhalation exposure chambers developed at PNL (U.S. Patent No. 4,216,741, August 12, 1980; Moss, 1980; Brown and Moss, 1981; Moss et al., 1982) and now commercially produced by the Harford System Division of Lab Products, Inc., Aberdeen, MD. The chamber (Figure 2) facilitates multiple-tier exposures of various laboratory animal species to aerosol- and vapor-laden atmospheres. The total volume of the chamber is 2.3 m^3 with an active mixing volume of 1.7 m^3 , the remainder being the inlet and exhaust volumes where animals are not placed. For rabbits, two

levels of caging were used, each level was split into two tiers which were offset from each other and from the chamber walls (Figure 2). Drawer-like stainless steel cage units composed of individual animal cages were suspended in the space above each tier. Stainless steel catch pans for the collection of urine and feces were suspended below each cage unit. The catch pans were left in position during each exposure period. Cageboard was added to these catch pans during non-exposure periods to reduce ammonia concentrations.

The chamber was designed so that uniform aerosol or vapor concentrations can be maintained throughout the chamber when the catch pans are left in position. Incoming air containing a uniform mixture of test material was diverted so that it flowed vertically along the inner surfaces of the chamber. Eddies were formed (Figure 1) at each tier as the aerosol or vapor flowed past the catch pans. Stagnant zones that would normally exist above each pair of catch pans were cleared by exhaust flow through the space between the tiers. Aerosol or vapor reaching the lowest level was deflected across the bottom tiers by metal strips in the space between the catch pan and the wall. Tests have shown that aerosol or vapor concentrations uniform to within 3 to 8% throughout the chamber can be obtained repeatedly provided the aerosol or vapor is uniformly mixed before passing through the chamber inlet (Moss 1980; Moss, et al. 1982). These tests were performed at PNL on a dynamically similar model of the chamber, as well as in the full scale chamber. Work at the Inhalation Toxicology Research Institute of the Lovelace Foundation, Albuquerque, NM has confirmed these findings (Griffis, et al. 1981).

Exposure Suite System Description

The chloroprene exposures were conducted using an automated data acquisition and control system in an exposure suite (Figure 3) consisting of three exposure rooms and a suite control center room (only one of the exposure rooms was used for chloroprene exposures). Data and comments from the exposure room were stored on separate magnetic disks and were printed on separate printers. Data were printed and stored immediately upon completion of the measurement to the "Daily Log". At the end of the day (24-hour period), the data were analyzed and a summary was printed. This summary included the mean, standard deviation, percent relative standard deviation,

maximum, minimum, and number of measurements for each set of data for the 24-hour period. A second printout provided a table of outliers (i.e. all data points which were beyond the critical limits defined in the protocol). A third printout provided a list of all comments generated by the computer and operators.

Generation and Delivery System

A schematic diagram of the chloroprene generation and delivery system is shown in Figure 4. The generator employed a rotary evaporation system (Büchi Rotavap, Model EL-131S, Brinkman Instruments Co., Westbury, NY) to produce high purity chloroprene vapor. A 1-liter evaporating flask was filled (under a nitrogen blanket) once at the beginning of each day with ~600 g of chloroprene from one or more of the original 1-gal glass containers, and was immersed in a hot water bath at ~150°F. A nitrogen stream was metered into the flask and flowed through the generator from the flask to the condenser, carrying chemical vapor to the condenser. The condenser, whose temperature was maintained by an ethylene glycol/water bath, brought the vapor to ~-2°C and condensed part of the total vapor. The condensation controlled the chloroprene vapor concentration produced by the generator and the refluxing action tended to remove low volatility impurities from the vapor.

From the condenser, the vapor entered a short distribution manifold from which individual delivery lines carried a metered amount of vapor to each exposure chamber. At the chamber end of each delivery line, an AirVac (AirVac Engineering Co., Milford, CN) pump withdrew the appropriate amount of chemical vapor from the manifold to reach the target concentration. Chamber concentrations were adjusted by the metering valves and/or by adjustment of the compressed air pressure to the vacuum pumps. At the end of the delivery line, the vapor entered a pneumatic 3-way valve where it was directed to the exposure chamber or the chamber exhaust system as appropriate. These valves allowed for a faster buildup of vapor concentration at start-up as well as more rapid diversion of chemical flow from chambers in the event rapid shutdown was necessary. At the start of a generation period, these valves were positioned to route the vapors to the chamber exhaust until the

concentrations in the distribution system had stabilized and volatile trace contaminants in the evaporation flask were purged. When the system was stable and exposures were ready to proceed, computer-controlled start-up of the exposure was initiated by rotation of the 3-way valves to the exposure position. Dilution of the nitrogen/chloroprene vapor mix with air to the target chamber concentrations occurred immediately before entry to the chambers.

Characterization of Test Article Concentration in Chambers

The buildup of vapor concentration at the beginning of exposure and the decay of concentration at the end of exposure are shown in Figure 5. Buildup and decay rates were measured prior to the start of the study without animals and during the study with animals to determine if the presence of animals would affect the buildup or decay rates. The time following the start of exposure for the concentration to reach 90% of the final stable concentration in the chamber (T_{90}), and the time following the termination of generation for the vapor concentration to decay to 10% of the stable concentration (T_{10}), were determined from the graphs (Table 1). Without animals present, the value of T_{90} was found to range from approximately 8 to 11 minutes. The value of T_{10} ranged from 8 to 10 minutes. At a chamber airflow rate of 15 air changes per hour, the theoretical value for T_{90} is approximately 12.5 minutes. With animals present in the chambers, values of T_{90} ranged from 9 to 11 minutes and the values for T_{10} were between 9 and 12 minutes. Contributing to the variations were slightly different flow rates in each chamber and variations in the discrete GC sample times relative to the exposure start and stop times. A T_{90} of 12 minutes was chosen for this study.

Uniformity of vapor concentration in the exposure chambers was measured prior to the start of the study and once during the study. During the prestart tests, uniformity measurements were made before the target concentrations were changed to the final levels. However, the measurements were not retaken as there was no indication of uniformity problems at the initial levels. The sample point was just above and about 10 cm in from the front or back center of each cage unit (Figure 6). The uniformity data for

each chamber is summarized in Table 2. Uniformity in all chambers was found acceptable (i.e., WPV≤5%, BPV≤5%, TPV≤7% as defined below).

The possible variation of test chemical concentration measured from one sample port to another during the measurement procedure is termed the total port variability (TPV) and consists of both spatial and temporal variations. Two factors contribute to the TPV. The first, the between port variability (BPV), is the factor of interest as it represents the spatial variation of test chemical distribution within the chamber. The second factor, the within port variability (WPV), represents the temporal fluctuation of the average chemical concentration within the chamber during the time the measurements were taken. This temporal factor includes variations in vapor generation as well as variation of the measurement instrument itself.

The WPV was determined from a minimum of three measurements taken at the on-line monitor port (1F or 2F) before, during and after all other ports were measured. The TPV was determined from measurements from, at the minimum, the front and back ports at each level on which animals were housed as well as one measurement from the on-line monitor port (whether or not animals were housed on that level). The BPV was determined by applying the following equation:

$$BPV = \sqrt{(TPV)^2 - (WPV)^2}$$

Since the WPV is often determined from fewer measurements than the TPV, statistically it is possible for the WPV to be greater than the TPV. In these cases, the BPV is very small, but it cannot be distinguished from the WPV. The BPV can't be determined using the above equation as it yields the square root of a negative number. In this case, it is reported as not resolvable.

Prior to the start of the study, before animals were placed in the chambers and again during the study with animals in the chambers, a Gardner Small Particle Detector (Type CN, Gardner Associates, Schenectady, NY) was used to check the room and all chambers for particles during generation. The minimum resolution of the Gardner counter is approximately 200 particles/cm³. No counts above the minimum resolvable level were measured in any chamber.

In order to determine the persistence of the chemical in the chamber following exposure, the concentration of chloroprene in one of the 175-ppm chambers was monitored following shutoff of the chemical flow to the chamber. Monitoring was performed once during the prestart activities without animals, and again during the study when animals were present (Figure 7). The concentration of chloroprene in the chamber without animals was below 1% of the initial concentration approximately 23 minutes following shutdown of the vapor generation system compared with approximately 24 minutes with animals in the chambers. The perturbation in the smooth decay at an elapsed time of approximately 35 minutes was due to opening of the chamber door to perform animal care activities.

Summaries of the concentration data during the study for all exposure chambers and the exposure room are included in Table 3. Summaries of concentration by exposure day are included in Appendix B along with graphic illustrations of the daily mean and standard deviation for each chamber. The means of concentrations in all chambers for the entire study were between 98 and 100% of the target, with relative standard deviations (%RSD) in the range of 1 to 2%. All individual concentration measurements were within $\pm 10\%$ of the target concentrations. The standard gas at the start of the study was produced from a diffusion-type generator with a target output of 4.6 ppm. After one day of exposure, the standard gas was switched to a pressurized gas cylinder with a concentration greater than 30 ppm.

The only significant excursion occurred on April 6, 1990 when exposure generation was terminated after five hours when a small leak in the generator accelerated the usage of chemical in the reservoir. A complete discussion of all concentration excursions and problems during exposures is included in Appendix B.

HEALTH AND SAFETY

This study was conducted under PNL's Health and Safety Plan. Protective clothing, including gloves and respiratory protection in the form of supplied-air respirators, was worn by all personnel working in the exposure room when exposure chamber doors were open. Personnel at risk from contacting liquid

chloroprene also wore Tyvek (EI duPont deNemours and Co., Wilmington, DE) outer protective clothing and appropriate chemical-resistant gloves.

The ACGIH TLV for chloroprene is 10 ppm. The PNL action limit is 1 ppm. During exposure periods, room concentration levels were monitored by a gas chromatograph. The highest concentration of chloroprene detected in the room was <MDL (MDL = 0.06 ppm). The lower limit of detection for the room monitor was <MDL. The results of room monitoring are presented in Appendix B. No personnel were exposed above the PNL action limit and no action was taken.

ANIMAL HUSBANDRY

New Zealand white rabbits (70 females; 7 males) were received in good condition on January 29, 1990 from R & R Rabbitry (Stanwood, WA). The females were 5-6 months old and the males were 6-7 months of age. Upon receipt the rabbits were housed in quarantine rooms 530 and 534 of the LSL-II building for ~8 weeks prior to the start of exposure. During the quarantine period males and females were housed separately, in individual cages on stainless steel wire racks equipped with automatic waterers. Inseminated females were acclimated in exposure chamber caging for three days prior to the start of exposures. Once exposures started chamber doors remained closed except during animal husbandry procedures. Rabbits were ~7-8 months of age at the beginning of exposure. Animals which were not selected for this study were discarded after the start of the first exposure.

The rabbits were examined by the veterinarian upon arrival, and all appeared to be in good condition. The veterinarian also examined the rabbits periodically during quarantine. During this time, two rabbits were treated with penicillin after being diagnosed with an upper respiratory infection. These rabbits were not used on the study. A control rabbit was sacrificed on the second exposure day because of cystitis and upper respiratory infection. *Pasteurella multocida* was recovered from the nasopharynx of 2 of the 3 rabbits with upper respiratory infections. The cause of the cystitis was not determined. Significant infection or other disease were not seen in any of the other rabbits. Animals were observed twice daily for mortality, morbidity, and clinical signs of toxicity during the study period.

A measured amount (≈ 170 g) of pelleted diet (Purina Certified Rabbit Chow #5322) was available from J type feeders during both the quarantine and exposure periods except during the daily chloroprene generation when the feed was removed. Food was discarded each day and new food was added. Contaminant analyses were performed by the manufacturer on each milling of diet. Samples of each milling used during the last 4 weeks of quarantine and during the exposure were collected at PNL and shipped to Lancaster Laboratories, Inc. (Lancaster, PA) for selected nutrient analyses. Animal drinking water, supplied by the City of Richland municipal system, was softened in PNL facilities and supplied to the animals *ad libitum* by automatic waterers. A representative sample of water from the LSL II animal facility was analyzed for contaminants during the exposure period. Rooms were illuminated by fluorescent lights with a 12-hour light, 12-hour dark, electrically operated cycle; light started at 0600.

Airflow in the chambers was maintained by the vacuum in the central chamber exhaust duct. Chamber airflow was measured by a multiplexed orifice-meter system consisting of a calibrated orifice located in each chamber exhaust, a Validyne Model DP-45 pressure transducer, a Validyne Model CD-18 carrier demodulator, and a Validyne Model PM-12 digital voltmeter. Airflow was automatically measured at ≈ 3 -hour intervals throughout each 24-hour day. Each flow orifice was calibrated prior to the start of the study to within ± 0.5 cfm.

Temperatures of the exposure chambers and the exposure room were measured by resistance temperature detectors (RTDs). The RTDs were placed in a representative location in each chamber (a top sample port on the back side). Each RTD was connected to an Omega Model 412B digital thermometer (Omega Engineering, Inc., Stamford, CT) by either a manual select switch or by computer controlled scanner relays. Temperatures were automatically recorded at ≈ 3 -hour intervals during each 24-hour day. RTDs were calibrated prior to the start of the study to $\pm 0.5^\circ\text{F}$.

Relative humidity was measured using an EG&G Model 910 chilled-mirror dewpoint hygrometer located in the exposure suite control center. Samples of the air from each measurement location were continuously pulled through polytetrafluoroethylene (PTFE) tubing to a central location in the exposure

suite control center. This assured a fresh sample at the dewpoint hygrometer. Air from the exposure chambers was sampled from a representative location (a top port on the back side). Percent RH was automatically recorded at regular intervals during the 24-hour day. Once the dewpoint had been determined by the hygrometer, the %RH was automatically calculated by the exposure system executive computer using the dewpoint value and the temperature (measured simultaneously at the same location by the RTD system). The following equation was used for this calculation:

$$\%RH = \frac{10^{\left[9.91 - \frac{2714.55}{(5/9)(T_1 - 32) + 293.13}\right]}}{10^{\left[9.91 - \frac{2714.55}{(5/9)(T_2 - 32) + 293.13}\right]}} \times 100$$

Where: T_1 = dewpoint temperature ($^{\circ}F$)
 T_2 = drybulb temperature ($^{\circ}F$)

Summations of chamber temperature, %RH, and airflow, for the entire study are shown in Table 4. This table includes the mean, the standard deviation (SD), mean expressed as a percentage of the target, the percent relative standard deviation (%RSD = $100 \times SD/\text{mean}$), the maximum and minimum values, number of samples, and the percent of samples for which the value was within the specified operating range.

The mean temperature values in all chambers for the entire study were between 66.6 and 68.2 $^{\circ}F$, all within the specified limits of 63 to 73 $^{\circ}F$. At least 86% of temperature readings for each chamber were within the specified limits. Extremes of the individual measurements ranged from 60.7 to 70.2 $^{\circ}F$. Critical low operating limit excursions, i.e. excursion of temperature $\leq 63^{\circ}F$, occurred during the first two days of animals in the chambers when the room temperature was set low to insure that chamber temperatures with the animal load did not exceed the high temperature limit.

The mean values of %RH in all chambers for the entire study ranged between 54.1 and 62.1% RH, all within the specified limits of 40 to 70%. At least 90% of all relative humidity readings for each chamber were within the

specified limits throughout the study. Extremes of the individual measurements (considering all chambers) ranged from 38 to 77%.

The mean values of chamber airflow in all chambers for the entire study were between 14.7 and 15.1 cfm (1 cfm = $\approx$ 1 air change per hour), all within the specified limits of 12 to 18 cfm. All (100%) of the measurements were within the specified limits.

A summary of the daily chamber environmental data and explanations of excursions of environmental data are provided in the exposure operation discussion sheets included in Appendix B.

DEVELOPMENTAL TOXICITY STUDY DESIGN

The developmental toxicity study in rabbits was comprised of the following treatment groups:

	Number of Rabbits		Number of Exposure Concentrations	Total
0 ppm	16	x	1	16
10 ppm	16	x	1	16
40 ppm	15	x	1	15
175 ppm	16	x	1	16
				63

Female rabbits were weighed 7 weeks after receipt. They were individually eartagged prior to arrival, and the eartag number is referred to as a pretest animal number. A study number was assigned to each rabbit (1-70) to acquire weight data using the XYBION PATH/TOX System (XYBION, Medical Systems Corp., Cedar Knolls, NJ).

Male rabbits were trained to serve an artificial vagina (AV) with the assistance of a teaser doe. The resulting semen sample was evaluated for motility. Acceptable samples from ≥ 3 bucks were pooled. A sperm count was performed and the semen was extended with oviduct stimulating medium to provide sufficient volume to inseminate ≈ 16 does. After the female rabbits were artificially inseminated, chorionic gonadotropin was injected in a marginal ear vein to promote ovulation.

The day female rabbits were artificially inseminated was designated as 0 days of gestation (dg). Bred females were weighed and randomly assigned to one of four exposure groups using body weight as the blocking variable. Insemination was conducted on three consecutive days (gestation groups A, B, and C). Following assignment to treatment groups the animals were individually caged, and on 3 dg were placed in exposure chambers for acclimation until the start of exposure on 6 dg. The disposition summary of study animals is shown in Table 5.

Inseminated female rabbits were exposed to chloroprene in whole-body exposure chambers at target concentrations of 0, 10, 40 and 175 ppm for 6 hours + T₉₀/day, 7 days/week, for 23 consecutive days (6 through 28 dg). Rabbits in gestation group A began exposure on 3/26/90, and groups B and C began on 3/27/90 and 3/28/90, respectively.

Exposure concentrations were selected on the basis of previous developmental toxicity studies in rats (Koeter and Appelman 1980), and on the lack of apparent maternal toxicity at these concentrations during a test exposure on inseminated New Zealand white rabbits.

Body weights were acquired on 0, 6, 15, 22, and 29 dg. Animals were observed twice each day for signs of chemical toxicity, moribundity, and mortality, 7 days per week.

On the day of scheduled sacrifice (29 dg) rabbits were weighed and killed by a lethal injection of Nembutal via an ear vein in the order of ascending study number. (The sacrifice was conducted blind as to treatment group.) Rabbits in gestation groups A, B, and C were killed on 4/18, 19, and 20/90, respectively. Animals were examined for gross tissue abnormalities, and the maternal liver and kidneys were removed and weighed. The uterus was removed, weighed, and opened. The number, position, and status (live, resorbed [early or late], or dead) of implants were recorded for each gravid uterus. Placentas were examined and discarded unless abnormal in appearance. Apparently non-gravid uteri from bred females were stained with 10% ammonium sulfide to detect possible implantation sites. Ovarian corpora lutea were counted for gravid females.

Live fetuses were weighed and examined for gross defects after a lethal injection of Nembutal, their sex was determined by internal examination of the gonads. All live fetuses in the uterus of each litter were examined for visceral defects by dissection of fresh tissue (modified from the method of Staples 1974). Fetal liver and kidney weights were obtained. The heads of fifty percent of the live fetuses were removed and placed in Louin's fixative. The first live fetus in the uterus to be selected for head removal was determined by a coin-toss, fetus #1 or #2, and the remainder were chosen alternately from the first. After fixation in Bouin's fluid, the heads were serially sectioned with a razor blade and examined for soft-tissue craniofacial abnormalities. All fetal carcasses, with and without heads, were prepared for skeletal staining. Cartilage as well as ossified bone was visualized by double-staining with alcian blue and alizarin red S. The individual identity of each fetal specimen was maintained throughout the study.

DATA ANALYSIS

Means and standard deviations for animal data were calculated with SAS® statistical software on a VAX 11/780 computer. Mean fetal body weights, as the mean of litter means, were analyzed using the SAS General Linear Models (GLM) Procedure (SAS 1985) with an analysis of variance (ANOVA) model for unbalanced data. Response variables, either body weight or the arc sine transformations of proportional incidence data, were analyzed against the class variable, "treatment", in a one-way ANOVA model. A Tukey's t-test (two-tailed) was used to assess statistically significant differences between control and exposed groups. If appropriate, the dose-response relationship was determined by means of an orthogonal trend test on arc sine transformed variables (Winer 1971). The litter was used as the basis for analysis of fetal variables.

DATA STORAGE

All residual animal tissues are stored in the LSL-II building, room 1428. All raw data and the study report are stored in the LSL-II building, rooms 1428 and 1229. The duration of storage will be in compliance with 21 CFR 58.195, or until NTP requests transfer of the data, whichever occurs first.

RESULTS

The chemical stability of chloroprene was evaluated in both the generator reservoir and in the exposure chambers (with and without animals). Chloroprene was found to be stable in the generator reservoir, and there was no evidence of degradation products greater than 0.1% of the target concentration in the exposure chambers (with or without animals). Direct measurements of chamber concentrations of BHT and peroxide confirmed that chamber concentrations of these two less volatile species were insignificant. Temperature-programmed GC failed to show evidence of any other degradation products.

The mean of the chloroprene concentration in each chamber for this study was between 98 and 100% of the target concentration, with relative standard deviations in the range of 1 to 2% (Table 3). The daily mean concentrations for all chambers were within 2% of the target concentrations (the daily protocol required the daily means to be within $\pm 10\%$ of the target concentrations). All individual concentration measurements were within $\pm 10\%$ of the target levels. The maximum concentrations observed in the control chamber and in the exposure room were less than the minimum detectable limit (0.06 ppm).

Each exposure group consisted of 15-16 artificially inseminated female rabbits which were killed on 29 dg. The overall pregnancy rate was 89% and ranged from 80 to 94% for each exposure group. One dam was found moribund due to an upper respiratory infection and cystitis and was killed on 7 dg. There were no maternal signs of toxicity attributable to exposure to the test material, although a few dams in each group exhibited nasal discharge, vaginal bleeding, and loose stools at various times during the study. A few dams in each treatment group were found to have minor lung lesions at the time of maternal sacrifice (Table 6); however, there was no apparent affect on their general health. The incidence of the clinical signs was low and dams appeared to be in excellent health throughout the study and at the time of sacrifice. There were no exposure-related effects on maternal body weight changes (Table 7). The negative adjusted maternal weight change observed in these animals is normal for mature rabbits during pregnancy.

Exposure of pregnant rabbits to chloroprene vapors on 6-28 dg had no effect on the number of implantations, the mean percent of live pups per litter, or on the percent of resorptions per litter (Table 8). Fetal body, kidney, and liver weights (as means of litter means) were not affected by gestational exposure to chloroprene, nor was the fetal sex ratio affected (Table 9).

The incidence of fetal malformations was not increased by exposure to chloroprene (Tables 10 and 12), in fact the incidence of malformations in the group as a whole was very low. There were no significant alterations in the incidence of total fetal variations or reduced ossifications among exposed groups (Tables 11 and 12). The variation termed "accessory blood vessel" refers to small extra vessels branching from the innominate or right subclavian arteries and would not be expected to affect the offspring. Another variation observed was termed "unusual lung lobes", this term refers to either missing or bifurcated diaphragmatic or cardiac lung lobes and would not be expected to affect the well-being of the offspring. This variation occurred only in exposed groups; however, the incidence was low (Table 12). Since variations in lung structure were noticed (but not recorded) in some of the dams, these variations may have been congenital and thus, not related to treatment. A review of the literature did not reveal any sources addressing the background incidence of variations in the lobulation of lungs in the New Zealand white rabbit.

DISCUSSION

Results of this study indicate that gestational exposure of New Zealand white rabbits to 10, 40, or 175 ppm chloroprene did not result in observable toxicity to either the dam or the offspring. The rabbit appears to be less sensitive to the effects of gestational exposure to chloroprene than the rat. Koeter and Appelman (1980) reported a reduction in fetal weight in Wistar rats at 175 ppm chloroprene, but the mean body weight of the rabbit pups was not affected at that concentration. Furthermore, although there was an increase in the incidence of resorptions at 175 ppm in the rat, this effect was not reproduced in the rabbit.

The highest exposure group, 175 ppm, was the no observable adverse effect level (NOAEL) for rabbits in the study with respect to both maternal and developmental toxicity.

REFERENCES

- Bartsch H., C. Malaveille, A. Barbin and G. Planche. 1979. Mutagenic and alkylating metabolites of halo-ethylenes, chlorobutadienes and dichlorobutenes produced by rodent or human liver tissues: Evidence for oxirane formation by P450-linked microsomal monooxygenases. Arch. Toxicol. 41(4):249-77.
- Bernstein, D.M. and R.T. Drew. 1980. The major parameters affecting temperature inside inhalation chambers. Am. Ind. Hyg. Assoc. J. 41:420-426.
- Brown, M.G. and O.R. Moss. 1981. An inhalation exposure chamber designed for animal handling. Lab. Anim. Sci. 31:717-720.
- Culik R., D.P. Kelly and J.J. Clary. 1978. Inhalation studies to evaluate the teratogenic and embryotoxic potential of β -chloroprene (2-chlorobutadiene-1,3). Toxicol. Appl. Pharmacol. 44:81-88.
- Gooch, J.J. and W.F. Hawn. 1981. Biochemical and hematological evaluation of chloroprene workers. J. Occup. Med. 23(4):268-272.
- Griffis, L.C., R.K. Wolff, R.L. Beethe, et al. 1981. Evaluation of a multi-Tiered inhalation chamber. Fundam. Appl. Toxicol. 1:8-12.
- Infante P.F., J.K. Wagoner and R.J. Young. 1977. Chloroprene: Observations of carcinogenesis and mutagenesis. In: Origins of Cancer, Cold Spring Harbor Laboratory.
- International Agency for Research on Cancer (IARC). 1979. Working Group on the Evaluation of the Carcinogenic Risk of Chemicals to Man. IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man. Vol. 19. World Health Organization, Geneva, Switzerland.
- Koeter, H.B.W.M. and L.M. Appelman. 1980. Inhalation embryotoxicity/teratogenicity studies with β -chloroprene in rats. Report No. R6387. Centraal Instituut voor Voedingsonderzoek, Utrechtseweg 48, Zeist.

- Moss, O. R. 1980. A chamber providing uniform concentration of particulates for exposure of animals on tiers separated by catch pans. In Workshop on Inhalation Chamber Technology, BNL 51318, Drew, R.T., editor. Brookhaven National Laboratory, Associated Universities, Inc., Upton, NY.
- Moss, O.R., J.R. Decker and W.C. Cannon. 1982. Aerosol mixing in an animal exposure chamber having three levels of caging with excreta pans. *Am. Ind. Hyg. Assoc. J.* 43:244-249.
- Plunkett, E.R. 1987. Handbook of Industrial Toxicology. 3rd Ed. Chemical Publishing Co., New York, NY.
- Salnikova, L.S. 1968. Embryotropic effect of volatile substances from nairit latexes. *Toksikol. Nov. Prom. Khim. Veshchestv.* 11:106-111.
- Salnikova, L.S. and V.N. Fomenko. 1973. Effect of chloroprene on embryogenesis. *Gig. Tr. Prof. Zabol.* 8:23-26.
- Sittig, M. 1985. Handbook of Toxic and Hazardous Chemicals. 2nd Ed. Noyes Publications, Park Ridge, NJ.
- Summer K.H. and H. Greim. 1980. Detoxification of chloroprene (2-chloro-1,3-butadiene) with glutathione in the rat. *Biochem. Biophys. Res. Commun.* 96(2):566-573.
- von Oettingen, W.F., W.C. Hueper, G. Deichmann, and F.M. Wiley. 1936. 2-Chlorobutadiene (chloroprene): Its toxicity and pathology and the mechanism of its action. *J. Ind. Hyg. Tox.* 18:240-270.

STREAM SELECT PORT ASSIGNMENTS

1	Not Used
2	175 ppm A
3	175 ppm B
4	40 ppm A
5	40 ppm B
6	10 ppm A
7	10 ppm B
8	Filtered Air
9	Room
10	0 ppm A
11	0 ppm B
12	Standard Gas

FIGURE 1. Chloroprene Inhalation Developmental Toxicity Study: Chamber Concentration Monitoring System

Moss et al. 1982. Amer. Ind. Hyg. Assoc. J. 43(4)244-249

FIGURE 2. Chloroprene Inhalation Developmental Toxicity Study: Inhalation Exposure Chamber U.S. Patent 4,216,741 (Top: Oblique Cutaway View; Bottom: Airflow Patterns)

FIGURE 3. Chloroprene Inhalation Developmental Toxicity Study: Study Exposure Suite

FIGURE 4. Chloroprene Inhalation Developmental Toxicity Study: Vapor Generation and Distribution System

FIGURE 5. Chloroprene Inhalation Developmental Toxicity Study: Buildup and Decay Curves of Concentration in Animal Chambers

FIGURE 5 (cont). Chloroprene Inhalation Developmental Toxicity Study:
Buildup and Decay Curves of Concentration in Animal Chambers

FIGURE 6. Chloroprene Inhalation Developmental Toxicity Study: Animal Exposure Chamber Schematic Showing Approximate Sampling Locations (F = Front; B = Back)

FIGURE 7. Chloroprene Inhalation Developmental Toxicity Study: Post-Exposure Monitoring Results of the 175 ppm A Chloroprene Chamber. (Persistence of the chemical in the chamber following exposure of animals.)

TABLE 1. Chloroprene Inhalation Developmental Toxicity Study: Summary of T₉₀^a and T₁₀ Data.

Target Concentration (ppm)	T ₉₀ [min]		T ₁₀ [min]	
	Prestart	Poststart	Prestart	Poststart
10 A ^b	10	10	10	12
10 B	8	10	8	10
40 A	9	11	10	9
40 B	11	11	10	10
175 A	10	11	9	10
175 B	9	9	10	9

^a A value of 12 minutes was used for T₉₀ for the study.

^b Two chambers were used (A and B) for each exposure concentration.

TABLE 2. Chloroprene Inhalation Developmental Toxicity Study:
Summary of Chamber Uniformity Data (On-line Measurements
Only).

Target Conc. (ppm)	TPV [%RSD]		WPV [%RSD]		BPV [%RSD]	
	Prestart	Poststart	Prestart	Poststart	Prestart	Poststart
10 A	0.6	1.3	0.1	0.8	0.6	1.0
10 B	0.2	0.4	0.2	0.2	0.2	0.4
40 A	0.5	0.6	0.7	0.1	--a	0.6
40 B	1.6	0.7	0.3	0.4	1.6	0.6
175 A	0.4	0.5	0.9	0.2	--a	0.4
175 B	0.1	0.5	0.2	0.2	--a	0.4

TPV = Total Port Variation. Acceptable limit $\leq 7\%$ RSD.

WPV = Within Port Variation. Acceptable limit $\leq 5\%$ RSD.

BPV = Between Port Variation. Acceptable limit $\leq 5\%$ RSD.

^a When the WPV is greater than the TPV, the BPV is very small and it cannot be resolved from the WPV.

Note: Concentrations in the chambers during the prestart measurements were at the originally proposed levels.

TABLE 3. Chloroprene Inhalation Developmental Toxicity Study: Summation of Exposure Concentration Data.

Target Concentration (ppm) ^a	Mean $\pm$ SD	Percent of Target $\pm$ %RSD	Maximum	Minimum	Number of Samples	Number of Samples In Range	Percent of Samples In Range
Room	<MDL ^b	—	<MDL ^b	<MDL ^b	272	—	—
0 A ^c	<MDL ^b	—	<MDL ^b	<MDL ^b	261	—	—
0 B	<MDL ^b	—	<MDL ^b	<MDL ^b	261	—	—
10 A	9.82 $\pm$ 0.18	98 $\pm$ 2	10.2	9.2	202	202	100
10 B	9.95 $\pm$ 0.22	100 $\pm$ 2	10.5	9.3	202	202	100
40 A	39.5 $\pm$ 0.69	99 $\pm$ 2	40.8	37.3	192	192	100
40 B	39.6 $\pm$ 0.84	99 $\pm$ 2	42.4	37.5	201	201	100
175 A	175 $\pm$ 2.45	100 $\pm$ 1	187	169	184	184	100
175 B	175 $\pm$ 1.62	100 $\pm$ 1	181	170	184	184	100
Std Gas ^d	4.3 $\pm$ 0.05	93 $\pm$ 1	4.3	4.2	9	9	100
Std Gas ^e	33.4 $\pm$ 1.79	111 $\pm$ 5	36	31.7	44	27	61
Std Gas ^f	36.3 $\pm$ 0.60	110 $\pm$ 2	37.8	33.1	57	25	44
Std Gas ^g	36.9 $\pm$ 0.40	102 $\pm$ 1	38.2	36.2	120	120	100

^a Acceptable Range = Target $\pm$ 10%

^b MDL (Minimum Detectable Limit = 0.06 ppm); QL (Quantifiable Limit = 0.20 ppm)

^c Two chambers were used (A and B) for each exposure concentration.

^d Standard in effect from 3/26/90 to 3/26/90. Target Concentration 4.6 ppm

^e Standard in effect from 3/27/90 to 3/31/90. Target Concentration 30 ppm

^f Standard in effect from 4/1/90 to 4/7/90. Target Concentration 33 ppm

^g Standard in effect from 4/8/90 to 4/19/90. Target Concentration 36.3 ppm

TABLE 4. Chloroprene Inhalation Developmental Toxicity Study: Summation of Chamber Environmental Data.

TEMPERATURE (°F)^a

Target Concentration (ppm)	Mean±SD	Percent of Target±%RSD	Maximum	Minimum	Number of Samples	Number of Samples In Range	Percent of Samples In Range
Room	64.2±1.8	99±3	67.9	58.5	203	195	96
0 A ^b	67.3±0.8	98±1	69.3	65.2	165	165	100
0 B	67.1±1.0	97±2	69.7	62.9	192	190	99
10 A	66.9±1.8	97±3	70.0	60.7	190	169	89
10 B	67.6±0.8	98±1	69.4	65.2	164	164	100
40 A	66.6±1.9	97±3	70.1	61.3	187	161	86
40 B	67.5±1.1	98±2	69.3	64.7	163	161	99
175 A	68.0±1.0	99±2	69.9	64.3	186	186	100
175 B	68.2±1.0	99±1	70.2	65.7	161	161	100

^aAcceptable Range = 65 to 73°F for the exposure chambers and 61 to 69°F for the exposure room.

^bTwo chambers were used (A and B) for each exposure concentration.

RELATIVE HUMIDITY (%RH)^c

Target Concentration (ppm)	Mean±SD	Percent of Target±%RSD	Maximum	Minimum	Number of Samples	Number of Samples In Range	Percent of Samples In Range
0 A	58.7±5.6	107±10	69	46	183	183	100
0 B	62.1±6.8	113±11	75	42	204	184	90
10 A	58.6±8.1	107±14	77	43	205	186	91
10 B	56.8±6.2	103±11	71	44	182	181	99
40 A	56.2±7.9	102±14	75	38	202	192	95
40 B	58.8±6.4	107±11	76	44	179	175	98
175 A	55.3±7.2	101±13	71	40	198	197	99
175 B	54.1±6.5	98±12	74	39	173	171	99

^cAcceptable Range = 40 to 70%.

AIRFLOW (CFM)^d

Target Concentration (ppm)	Mean±SD	Percent of Target±%RSD	Maximum	Minimum	Number of Samples	Number of Samples In Range	Percent of Samples In Range
0 A	14.7±0.4	98±3	15.5	13.2	175	175	100
0 B	14.8±0.4	99±3	17.4	13.5	198	198	100
10 A	14.8±0.2	99±2	15.8	13.2	198	198	100
10 B	14.8±0.3	99±2	15.3	12.7	174	174	100
40 A	14.8±0.2	99±1	15.2	13.1	198	198	100
40 B	15.0±0.3	100±2	15.7	13.1	175	175	100
175 A	15.0±0.3	100±2	15.7	13.1	198	198	100
175 B	15.1±0.3	100±2	15.7	13.3	175	175	100

^dAcceptable Range = 12 to 18 CFM.

TABLE 5. Chloroprene Inhalation Developmental Toxicity in Rabbits: Study Disposition.

Target Chloroprene Concentration (ppm)	Treatment Group	Inseminated	Pregnant	Removed
0	1	16	12	1 ^a
10	2	16	15	0
40	3	15	13	0
175	4	16	15	0

^a One dam was found moribund on 3/27/90 (7 dg) leaving 15 inseminated does in this group.

TABLE 6. Chloroprene Inhalation Developmental Toxicity Study. Summary of Gross Observations at Time of Maternal Sacrifice.

Target Chloroprene Concentration (ppm)	0	10	40	175
N	15	16	15	16
Lungs:				
Khaki colored foci	1	1	0	1
Dark red foci	0	0	0	1
Granular tissue	0	1	1	0
Cyst (3-4 mm)	1	0	0	0
Liver Lesions:				
Yellow (1 mm)	0	0	1	0
Pale (2x4 mm)	1	0	0	0
Bladder:				
Thickened wall	0	0	0	1

TABLE 7. Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Mean Body, Uterine, Extra-gestational Gain^a and Organ Weights of Pregnant Developmental Toxicology Rabbits (g $\pm$ SD).

Target Chloroprene Concentration (ppm)	0	10	40	175
N	12	15	13	15
Body Weight				
0 dg	4359 $\pm$ 202	4374 $\pm$ 277	4399 $\pm$ 208	4344 $\pm$ 253
6 dg	4377 $\pm$ 206	4340 $\pm$ 237	4391 $\pm$ 176	4373 $\pm$ 270
15 dg	4467 $\pm$ 212	4397 $\pm$ 237	4508 $\pm$ 169	4463 $\pm$ 294
22 dg	4525 $\pm$ 236	4458 $\pm$ 250	4569 $\pm$ 222	4545 $\pm$ 322
29 dg	4607 $\pm$ 200	4534 $\pm$ 318	4641 $\pm$ 283	4655 $\pm$ 328
Adjusted Maternal Weight Change	-219 $\pm$ 148	-205 $\pm$ 213	-141 $\pm$ 167	-132 $\pm$ 182
Uterine	467 $\pm$ 120	365 $\pm$ 211	384 $\pm$ 158	443 $\pm$ 156
Liver	122 $\pm$ 24	119 $\pm$ 27	135 $\pm$ 30	122 $\pm$ 22
Percent LBWR^b	2.7 $\pm$ 0.5	2.6 $\pm$ 0.5	2.9 $\pm$ 0.6	2.6 $\pm$ 0.4
Kidney	20.5 $\pm$ 3.0	18.8 $\pm$ 2.2	20.5 $\pm$ 2.2	19.2 $\pm$ 2.4
Percent KBWR^c	0.4 $\pm$ 0.1	0.4 $\pm$ 0.0	0.4 $\pm$ 0.0	0.4 $\pm$ 0.0

^a Adjusted Maternal Body Weight Change = body weight (20 dg) - body weight (0 dg) - uterine weight.

^b LBWR = liver to body weight ratio $\times$ 100.

^c KBWR = kidney to body weight ratio $\times$ 100.

TABLE 8. Chloroprene Inhalation Developmental Toxicity Study: Reproductive Measures (Mean $\pm$ SD) in Rabbits.

Target Chloroprene Concentration (ppm)	0	10	40	175
NUMBER OF:				
Inseminated Females	15	16	15	16
Number Pregnant	12	15	13	15
Pregnancies Examined	12	15	13	15
Corpora Lutea/Dam	12.4 $\pm$ 3.6	8.9 $\pm$ 4.1	10.5 $\pm$ 3.7	11.1 $\pm$ 3.7
Implantations/Dam	8.9 $\pm$ 2.7	7.1 $\pm$ 3.4	7.0 $\pm$ 3.0	8.0 $\pm$ 2.3
Live Fetuses/Litter	7.8 $\pm$ 2.6	5.8 $\pm$ 3.6	6.2 $\pm$ 2.9	7.0 $\pm$ 2.6
Resorptions/Litter:	1.2 $\pm$ 1.3	1.3 $\pm$ 1.9	0.8 $\pm$ 1.0	1.0 $\pm$ 1.1
Early	0.7 $\pm$ 1.0	1.1 $\pm$ 1.9	0.4 $\pm$ 0.8	0.7 $\pm$ 1.2
Late	0.5 $\pm$ 0.8	0.1 $\pm$ 0.4	0.4 $\pm$ 0.9	0.3 $\pm$ 0.5
Dead Fetuses/Litter	0	0	0	0
Litters with Resorptions	7	9	6	10
Litters with $\geq$ 2 Resorptions	4	3	3	3
PERCENTAGE OF:				
Pregnant Females	80	94	87	94
Live Fetuses/Litter	87.2 $\pm$ 13.7	72.7 $\pm$ 36.2	82.9 $\pm$ 28.1	84.8 $\pm$ 24.5
Resorptions/Litter:	12.8 $\pm$ 13.7	27.3 $\pm$ 36.2	17.1 $\pm$ 28.1	15.2 $\pm$ 24.5
Early	8.2 $\pm$ 12.3	25.3 $\pm$ 37.3	11.3 $\pm$ 28.1	12.0 $\pm$ 25.5
Late	4.6 $\pm$ 7.3	2.0 $\pm$ 5.6	5.8 $\pm$ 12.0	3.3 $\pm$ 5.8
Dead Fetuses/Litter	0	0	0	0
Litters with Resorptions	58	60	46	67
Litters with $\geq$ 2 Resorptions	33	20	23	20

TABLE 9. Chloroprene Inhalation Developmental Toxicity Study: Fetal Organ and Body Weights (grams), Fetal Organ to Body Weight Ratio, and Fetal Sex Ratio in Rabbits (Mean of Litter Means; $\bar{x} \pm SD$).

Target Chloroprene Concentration (ppm)	0	10	40	175
Litters Examined	12	13	12	14
Fetal Weight	43.3 $\pm$ 6.2	45.8 $\pm$ 5.5	42.0 $\pm$ 4.9	43.8 $\pm$ 3.7
Male	42.2 $\pm$ 6.9	46.0 $\pm$ 5.2	41.8 $\pm$ 5.8	44.0 $\pm$ 5.0
Female	44.2 $\pm$ 5.7	45.5 $\pm$ 6.1	42.0 $\pm$ 5.0	43.6 $\pm$ 4.1
Fetal Liver Weight	3.0 $\pm$ 0.7	3.2 $\pm$ 0.5	2.9 $\pm$ 0.5	3.1 $\pm$ 0.4
Male	2.9 $\pm$ 0.8	3.3 $\pm$ 0.5	2.9 $\pm$ 0.5	3.1 $\pm$ 0.5
Female	3.1 $\pm$ 0.6	3.3 $\pm$ 0.6	2.9 $\pm$ 0.6	3.1 $\pm$ 0.4
Percent LBWR ^a	6.9 $\pm$ 0.6	7.0 $\pm$ 0.4	7.0 $\pm$ 0.6	7.0 $\pm$ 0.4
Male	6.8 $\pm$ 0.8	7.1 $\pm$ 0.5	7.0 $\pm$ 0.5	7.1 $\pm$ 0.6
Female	6.9 $\pm$ 0.5	7.1 $\pm$ 0.5	6.9 $\pm$ 0.7	7.0 $\pm$ 0.5
Fetal Kidney Weight	0.37 $\pm$ 0.05	0.41 $\pm$ 0.15	0.35 $\pm$ 0.05	0.37 $\pm$ 0.03
Male	0.36 $\pm$ 0.07	0.43 $\pm$ 0.18	0.36 $\pm$ 0.06	0.37 $\pm$ 0.05
Female	0.38 $\pm$ 0.05	0.37 $\pm$ 0.06	0.35 $\pm$ 0.06	0.37 $\pm$ 0.03
Percent KBWR ^b	0.85 $\pm$ 0.06	0.90 $\pm$ 0.28	0.84 $\pm$ 0.06	0.84 $\pm$ 0.03
Male	0.86 $\pm$ 0.06	0.93 $\pm$ 0.34	0.86 $\pm$ 0.10	0.84 $\pm$ 0.05
Female	0.86 $\pm$ 0.06	0.82 $\pm$ 0.06	0.83 $\pm$ 0.06	0.84 $\pm$ 0.04
Percent Male Fetuses	52 $\pm$ 24	57 $\pm$ 21	50 $\pm$ 11	49 $\pm$ 16

^a LBWR = liver to body weight ratio $\times$ 100.

^b KBWR = kidney to body weight ratio $\times$ 100

TABLE 10. Chloroprene Inhalation Developmental Toxicity Study:
Malformations Observed in Live Rabbit Fetuses.

Target Chloroprene Concentration (ppm)		Fetuses ^a				Litters ^a			
		0	10	40	175	0	10	40	175
Total Examined ^b		93	87	81	105	12	13	12	14
Heads examined ^c		47	45	43	55	12	13	12	14
Skulls examined ^d		93	87	81	105	12	13	12	14
Viscera examined ^e		93	87	81	105	12	13	12	14
MALFORMATIONS:									
Hydrocephaly	No. (%)	-	-	1 (2.3)	-	-	-	1 (8.3)	-
Limb Flexure	No. (%)	1 (1.1)	-	1 (1.2)	-	1 (8.3)	-	1 (8.3)	-
TOTAL:									
Malformations	No.	1	0	2	0	-	-	-	-
Fetuses (Litters) with Malformations	No. (%)	1 (1.1)	0 (0.0)	2 (2.5)	0 (0.0)	1 (8.3)	0 (0.0)	2 (16.7)	0 (0.0)

^a A single fetus or litter may be represented more than once in this table.

^b All fetuses examined for external and skeletal defects. One-half had heads removed prior to skeletal staining.

^c Heads fixed in Bouin's solution for evaluation of soft-tissue craniofacial evaluations.

^d Heads remained on the fetuses for skeletal examination; see (b).

^e Visceral examinations performed on 100% of live fetuses.

TABLE 11. Chloroprene Inhalation Developmental Toxicity Study: Variations and Reduced Ossifications Observed in Live Rabbit Fetuses.

Target Chloroprene Concentration (ppm)		Fetuses ^a				Litters ^a			
		0	10	40	175	0	10	40	175
Total Examined ^b		93	87	81	105	12	13	12	14
Heads examined ^c		47	45	43	55	12	13	12	14
Skulls examined ^d		93	87	81	105	12	13	12	14
Viscera examined ^e		93	87	81	105	12	13	12	14
VARIATIONS:									
Supernumerary:									
Rib	No.	55	48	46	50	12	13	11	13
	(%)	(59.1)	(55.2)	(56.8)	(47.6)	(100)	(100)	(91.7)	(92.9)
Rudimentary Rib	No.	13	16	16	26	8	7	9	11
	(%)	(14.0)	(18.4)	(19.8)	(24.8)	(66.7)	(53.8)	(75.0)	(78.6)
Ossification Site	No.	2	-	-	1	2	-	-	1
	(%)	(2.2)			(1.0)	(16.7)			(7.1)
Misaligned Sternebra	No.	2	-	-	2	2	-	-	2
	(%)	(2.2)			(1.9)	(16.7)			(14.3)
Accessory Blood Vessel ^f	No.	2	3	-	2	1	2	-	2
	(%)	(2.2)	(3.4)		(1.9)	(8.3)	(15.4)		(14.3)
Unusual Lung Lobe ^f	No.	-	4	4	6	-	2	2	4
	(%)		(4.6)	(4.9)	(5.7)		(15.4)	(16.7)	(28.6)
Short Tail	No.	-	1	3	-	-	1	2	-
	(%)		(1.1)	(3.7)			(7.7)	(16.7)	
Fused Sternebrae	No.	-	-	1	-	-	-	1	-
	(%)			(1.2)				(8.3)	
REDUCED OSSIFICATIONS:									
Pelvis	No.	-	-	1	1	-	-	1	1
	(%)			(1.2)	(1.0)			(8.3)	(7.1)
Sternebra	No.	36	25	17	37	9	9	8	11
	(%)	(38.7)	(28.7)	(21.0)	(35.2)	(75.0)	(69.2)	(66.7)	(78.6)
Vertebral Centra	No.	1	-	1	-	1	-	1	-
	(%)	(1.1)		(1.2)		(8.3)		(8.3)	
TOTAL:									
Reduced Ossifications and Variations ^g		No.	111	97	89	125	-	-	-
Fetuses (Litters) with Variations		No.	83	75	71	95	12	13	12
		(%)	(89.2)	(86.2)	(87.7)	(90.5)	(100)	(100)	(100)

^a A single fetus or litter may be represented more than once in this table.

^b All fetuses examined for external and skeletal defects. One-half had heads removed prior to skeletal staining.

^c Heads fixed in Bouin's solution for evaluation of soft-tissue craniofacial evaluations.

^d Heads remained on the fetuses for skeletal examination; see (b).

^e Visceral examinations performed on 100% of live fetuses.

^f See text for description of accessory blood vessel and unusual lung lobe.

^g There may be >1 variation per fetus.

TABLE 12. Chloroprene Inhalation Developmental Toxicity Study. Mean Percent of Live Rabbit Fetuses Affected per Litter (Mean Percent $\pm$ SD).

Target Chloroprene Concentration (ppm)	0	10	40	175
Litters Examined	12	13	12	14
MALFORMATIONS:				
Hydroencephaly	-	-	2.8 $\pm$ 9.6	-
Limb Flexure	0.9 $\pm$ 3.2	-	0.8 $\pm$ 2.9	-
Total Malformations	0.9 $\pm$ 3.2	-	2.2 $\pm$ 5.4	-
VARIATIONS:				
Supernumerary:				
Rib	63.1 $\pm$ 25.4	60.8 $\pm$ 25.5	56.3 $\pm$ 33.1	46.1 $\pm$ 27.5
Rudimentary Rib	13.2 $\pm$ 13.7	13.7 $\pm$ 15.1	21.1 $\pm$ 16.9	26.2 $\pm$ 19.0
Ossification Site:	1.7 $\pm$ 4.0	-	-	0.9 $\pm$ 3.3
Misaligned Sternebra	1.9 $\pm$ 4.3	-	-	2.1 $\pm$ 5.7
Accessory Blood Vessel ^a	1.9 $\pm$ 6.4	4.3 $\pm$ 10.7	-	2.3 $\pm$ 6.1
Unusual Lung Lobe ^a	-	2.9 $\pm$ 7.5	5.4 $\pm$ 13.0	7.1 $\pm$ 13.1
Short Tail	-	1.0 $\pm$ 3.5	4.4 $\pm$ 11.8	-
Fused Sternebrae	-	-	1.7 $\pm$ 5.8	-
REDUCED OSSIFICATION:				
Pelvis	-	-	1.2 $\pm$ 4.1	0.6 $\pm$ 2.4
Sternebra	33.8 $\pm$ 27.8	25.8 $\pm$ 28.2	19.8 $\pm$ 19.9	32.4 $\pm$ 29.0
Vertebral Centra	0.9 $\pm$ 3.2	-	1.0 $\pm$ 3.6	-
TOTAL VARIATIONS	89.1 $\pm$ 10.7	85.1 $\pm$ 18.0	87.2 $\pm$ 14.1	89.7 $\pm$ 10.6

^a See text for description of accessory blood vessel and unusual lung lobe.

APPENDIX A

CHEMISTRY MONITORING NARRATIVE AND DATA

BNW Bulk Chemical Reanalysis Report
Test Chemical Characterization, Storage, Usage and
Disposition
Test Chemical Concentration Monitoring
Test Chemical Stability in the Exposure System
GC/MS Analysis of Chloroprene and Its Dimer Products

BNW Bulk Chemical Reanalysis Report

BULK CHEMICAL REANALYSIS

COMPOUND: CHLOROPRENE
CAS# 126-99-8
LOT# BNW# 53436-20 (bottle #2)
VENDOR LOT# Not available
APPEARANCE: Colorless liquid
RECEIPT DATE: 2/14/90
ANALYSIS PERIOD: Initial
STORAGE TEMPERATURE: --20°C
SAMPLE SUBMITTAL DATE: 3/2/90
SAMPLE ANALYSIS DATE: 3/2/90
ANALYSIS PROCEDURE: Based on MRI Method, May 22, 1986
NOTEBOOK REFERENCE: BNW 53436-56

ASSAY: Gas chromatography used a DB-5 capillary column, 30 m X 0.53 mm
3/9/90 1.5 μ film thickness for purity by area percent.
Parameters included initial temperature of 35°C for 5 minutes with a
program rate of 10°C/minute and a final temperature of 275°C for 5 minutes.
Instrument: HP 5890

RESULTS:	<u>BULK PURITY</u>	<u>AVERAGE</u>	<u>RSD</u>
	98.49%		
	98.51%	98.51%	0.01%
	98.52%		

Chloroprene eluted at ~3.29 minutes.
Sample injections were allowed to run ~30 minutes past chloroprene peak to
determine if any impurities were present. Chlorobutene and cis/trans-1-
chlorobutadiene constitute the major impurities (about 0.6 and 0.1%
respectively). The major dimer eluted at ~17.06 minutes with 0.24% of total
chloroprene sample area.

IDENTITY: Infrared spectroscopy used silver chloride plates scanned from
3/2/90 4000 cm⁻¹ to 600 cm⁻¹.
Instrument: Beckman Aculab 8

RESULTS: The spectrum was similar to copy of MRI spectrum.

PEROXIDES: A Fisher Computer Aided Titrimeter was utilized for peroxide analysis.
3/7/90

Peroxide Meq/Kg	Average Meq/Kg	RSD
0.08		
0.05	0.06	34.8%

Conclusion: Infrared spectroscopy confirms the identity of lot BNW# 53436-20
(bottle #2). Gas chromatography shows this lot to be 98.51% pure by
area percent with the major dimer constituting 0.24% of the total
chloroprene sample area. The peroxide is within the limit of
1.0 Meq/Kg. Chloroprene lot BNW533436-20 is acceptable for use.

Signature of Technician: Neta Sue Stamps Date: 3-10-90

Reviewed by: N.B. Valentini Date: 3-12-90

Signature of Chemist: RBW Date: 3-12-90

**Test Chemical Characterization, Storage, Usage and
Disposition**

I. TEST CHEMICAL CHARACTERIZATION, STORAGE, USAGE AND DISPOSITION

A. Test Material Receipt, Storage and Usage

1. Receipt

Chloroprene test material was received from Denka Chemical Corporation, Houston, Texas 77017. Twelve, four-liter bottles containing a total of ~46 kg of chloroprene [BNW Lot No. 53436-20, bottles 1 through 12] were received 2/14/90. The test material was inhibited with ~ 0.1% each of phenothiazine and t-butylcatechol at Denka. The material was shipped at ambient temperature.

2. Storage Conditions

The bulk chemical was stored in its original container at ~ -20°C under a nitrogen headspace in the Chemical Storage and Transfer Facility adjacent to the LSL-II laboratory.

3. Usage

An average of 0.6 kg chloroprene was consumed per exposure day. The study required about 15 kg for animal exposure.

4. Transfer Procedures

A one liter evaporation flask was filled (under a nitrogen blanket) once at the beginning of each day with ~ 600 grams of chloroprene from one of the original four-liter glass containers. The flask was kept at a positive nitrogen atmosphere to prevent the introduction of air to the system during the generation period.

5. Waste Disposal

Excess used test material was transferred to a labelled glass bottle containing ~2 g each of phenothiazine and tert-butylcatechol in ~1 liter of xylene. Full bottles were stored at LSL-II until disposed of by the BNW Waste Management and Environmental Compliance Group.

6. Surplus Disposal

Surplus test material (e.g., unused bulk material) will be disposed of by the BNW Waste Management and Environmental Compliance Group at the end of all studies currently planned unless otherwise instructed.

B. Chemical Analysis

1. Analysis at Midwest Research Institute

Chemical characterization of chloroprene test material (supplied by Denka) was presented in the May 22, 1986 MRI Report and provides an indication of test material purity to be expected. Bulk chemical (BNW Lot No. 12103-4-1, MRI Batch No. 01) was identified as chloroprene. Cumulative analytical data indicated a purity of approximately 96% as received by MRI.

The observed spectra by infrared, ultraviolet/visible and nuclear magnetic resonance analyses were also consistent with literature references.

Elemental analysis results for carbon and hydrogen were in agreement with theoretical values. Karl Fischer titrimetry indicated less than 0.01% water. Analysis for peroxide by titration indicated 1.96 ± 0.06 meq/kg of test material. Inhibitor (t-butylcatechol) was approximated to be present at 40 to 50 ppm. It is important to note that the present material is inhibited to 0.1% (1000 ppm) with t-butylcatechol and phenothiazine.

Thin-layer chromatographic analysis indicated a major spot only. Gas chromatographic analyses by two systems each indicated impurities with total peak areas of 0.96% and 3.30% relative to the major peak area. Five impurities were tentatively identified as three chloroprene dimers, a chlorobutene, and a chloroprene isomer by gas chromatography/mass spectrometry.

An accelerated stability study (55°C for up to 6 hours) indicated that in addition to dimerization (~62% increase in dimer content), significant polymerization also occurs.

An unopened bottle of the bulk chemical was shipped to Research Triangle Institute on 2/20/90 for chemical characterization. The results of this assay were not available at the time of report preparation.

2. Reanalysis at Battelle Pacific Northwest Laboratories

The MRI recommended procedure, based on the May 22, MRI Special Report "Chemical Characterization of Chloroprene" was implemented as BNW SOP# 0B-AC-3A14. Bulk chemical purity was monitored upon receipt and at 16 $\pm$ 2 week intervals during storage and use at BNW. The bulk chemical reanalysis included an analysis within thirty days prior to the start of the study. Subsequent reanalysis of bulk material were performed using gas chromatography to determine purity (%) of chloroprene and dimer content. Analysis to determine peroxide content of each bottle was performed within 6 weeks prior to use as generation material.

Identity of bulk chemical was confirmed during initial bulk analysis by infrared spectroscopy. Bulk purity analysis was performed by gas chromatography using a DB-5 capillary column, 30 m x 0.53 mm i.d. fused silica, 1.5 μ film thickness. Concentrations of phenothiazine and t-butylcatechol (inhibitors) were verified upon receipt by gas chromatography. Test material BNW Lot No. 53436-20 bottle #2 was 98.51% pure upon receipt and was found to contain ~ 0.06 meq/kg of peroxides at the time of the analysis. The predominate dimer isomer was present at ~0.24%. Chlorobutene and 1-chlorobutadiene were present at ~0.6 and 0.1%, respectively. Thus, bulk purity was similar to that observed by BNW from past experience with chloroprene test material. However, the concentration of dimer was about 2 to 3 times greater than seen in the earlier repeated dose and subchronic studies performed at BNW. The greater dimer content may have resulted from shipment at ambient temperature. The earlier shipments were via refrigerated freight.

Test Chemical Concentration Monitoring

II. TEST CHEMICAL CONCENTRATION MONITORING

A. Monitoring System Description

Chamber and room concentrations of chloroprene were determined using an on-line Hewlett-Packard Model 5890 gas chromatograph equipped with a flame ionization detector operated at 250°C. A 1/8 inch O.D. x ~1 ft. nickel column packed with 1% SP1000 on 60/80 Carbopack B was used to separate chloroprene from other components. The carrier gas was nitrogen at a flow rate of ~30 ml/min. The column was maintained isothermally at 120°C. Under these conditions, chloroprene exhibited a retention time of ~0.66 minutes.

An oven-mounted, 12-port stream select valve constructed of Hastelloy-C provided the interface between the on-line gas chromatograph and the exposure chambers, the control chambers, the exposure room, the on-line standard and a filtered air blank. This valve directed a continuous stream of sampled atmosphere to a 1.0 ml nickel sample loop outfitted with an oven-mounted sampling valve (Hastelloy-C). Automatic switching of the stream select valve allowed access to all test chambers, the room, a blank and the on-line standard. A schematic diagram of the chamber concentration monitoring system is shown in Figure A.1. Individual 1/4-inch Teflon sample lines (also used for monitoring relative humidity) lead from each chamber to a position close to the on-line monitor. These sample lines had a flow of 2 to 2.5 l/min. Teed from each of these lines were 1/16-inch Teflon lines, ~4 feet in length, which lead to each port of the stream select valve. These lines were purged at ~30 ml/min for ~2.6 min prior to sample injection.

Data were transferred from the HP5890 chromatographic integrator to an HP85B computer. The HP85B computer remotely controlled the selection of the correct sample stream and the operation of the monitor. Each analysis required about 3 minutes. Thus, all chamber positions, the room, the blank, and the on-line standard were monitored approximately every 36 minutes. The equation of the calibration curve was contained in the HP85B and was applied to the data transmitted by the on-line GC monitor. The HP85B also accumulated and printed the sample values until all 12 ports of the stream select valve had been measured. These values were then sent to the executive computer for printing and storage. Each value accumulated by the HP85B was compared with limit values for that particular location. If the value was beyond the control limits, the HP85B immediately sent the information to the executive computer which took the appropriate action.

As described in the section that follows, the monitor was calibrated against gravimetrically prepared standards. The gravimetric standards were related to the on-line monitor through quantitative analysis of grab samples taken from exposure chambers simultaneously sampled by the on-line GC while the on-line GC was kept in normal rotation.

Additionally, the operation of the chamber monitor was checked throughout the day against an on-line standard. This check provided a measure of day-to-day instrument drift. Additional calibration checks by grab sampling were performed to check the monitor calibration when drift of the on-line standard response factor was detected.

Daily operating procedures for the concentration monitoring system are contained in SOP# 0B-AC-3B1N and 0B-BE-3B2V.

B. Calibration of the On-Line Monitor

The relationship between the on-line GC response and the concentration of chloroprene in the chamber was established from independent analysis of chamber grab samples taken directly from the exposure chambers during a routine exposure period. The calibration of the on-line monitor was established by correlation of the on-line monitor peak area (at the time of grab sampling) against independent measurement of chamber concentration (analysis of chamber grab samples). The normal dynamic operation of the monitor was not disrupted during collection of these grab samples.

Grab samples were obtained by collecting chloroprene in bubblers containing 20 ml of dimethylformamide (DMF). Known volumes of chamber atmosphere from each chamber were collected using a calibrated critical orifice controlled sampler at a constant flow rate of approximately 1.0 l/min. Internal standard solution was added to each bubbler after sample collection to correct for any volume change in the DMF, variations in injection volume, and instrument drift. Breakthrough of chloroprene was determined with a back-up bubbler linked in series to the primary bubbler used to sample one of the 175 ppm exposure chambers. Typical breakthrough for a 2 liter sample was approximately 5%. Total mass of chloroprene collected in the primary bubblers was corrected for breakthrough.

Grab samples and standards were analyzed on an off-line Hewlett-Packard gas chromatograph equipped with a flame ionization detector operated at 250°C and a glass column packed with 10% Carbowax 20M on 80/100 Chromasorb. One microliter sample volumes were injected using an on-column configuration and an automatic liquid sampler. The initial oven temperature (70°C) was held isothermally for 3 minutes. The temperature was then ramped at 10°C/minute to 150°C and held for 20 minutes. Chloroprene eluted at ~1.9 minutes and the dodecane internal standard at ~8.0 minutes using these parameters.

Gravimetrically prepared standard solutions of chloroprene in DMF were used to calibrate the off-line GC used to quantitate chloroprene collected by grab sampling. A set of four standards was run for each analysis session. The concentration range of the standards bracketed the expected concentration range of the chamber grab samples. Two independently weighed stock solutions were used each time the standard series was prepared. The preparation of dual stock solutions helped "flag" any weighing errors. A linear calibration curve with a zero (Figure A.2) intercept was used for the on-line monitor determination of chloroprene concentrations in the exposure chambers, room, air blank and on-line standard.

C. On-Line Standard

An on-line standard of chloroprene was used to check instrument drift throughout each exposure day. The on-line standard of chloroprene in nitrogen gave reproducible day-to-day results and within day results having relative standard deviations of <1% as shown in the table in section E. The data used to determine the reproducibility of the on-line standard was taken from three days each having 6 hours of continuous generation.

The chloroprene standard was monitored every 12th sample throughout the exposure period. It was also checked before the start of each exposure day. The measured concentration for the standard was required to be within $\pm 10\%$ of

STREAM SELECT PORT ASSIGNMENTS

- 1 Filtered Air
- 2 175 ppm A
- 3 175 ppm B
- 4 40 ppm A
- 5 40 ppm B
- 6 10 ppm A
- 7 10 ppm B
- 8 Filtered Air
- 9 Room
- 10 0 ppm A
- 11 0 ppm B
- 12 Standard Gas

FIGURE A.1. Chloroprene Inhalation Developmental Toxicity Study: Diagram of the Exposure Chamber Concentration Monitoring System.

Variable Name	Coefficient	Std. Err. Estimate	t Statistic	Prob > t
area ratio	5.748e+2	3.414e-1	1.684e+3	2.93e-18

Data File: Chloroprene Standards 3/5/90

Source	Sum of Squares	Deg. of Freedom	Mean Squares	F-Ratio	Prob>F
Model	2.608e+6	1	2.608e+6	1.480e+6	2.08e-17
Error	1.057e+1	6	1.762e+0		
Total	2.608e+6	7			

Coefficient of Determination (R^2) 1.000e+0
 Adjusted Coefficient (R^2) 1.000e+0
 Coefficient of Correlation (R) 1.000e+0
 Standard Error of Estimate 1.327e+0
 Durbin-Watson Statistic 1.728e+0

FIGURE A.2. Chloroprene Inhalation Developmental Toxicity Study: Calibration Curve for Gravimetrically Prepared Standards.

the assigned target value before any exposure could begin without consultation with the exposure control task leader. During the course of the exposure, if the on-line standard was within $\pm 5\%$ of the target value, no change in calibration was required. If the on-line standard was beyond $\pm 5\%$ of its assigned target, the chemist was informed immediately by an exposure specialist. Additional grab sampling was initiated by the chemist on a case by case basis when drift in the on-line standard was between $\pm 5-10\%$. A drift of 10% or more required high priority recalibration of the on-line monitor by grab sampling.

D. Detection Limits

1. Definition of MDL, MQL, MLQ and QL

The Minimum Detectable Limit (MDL) is defined as the lowest concentration of analyte that can be reliably detected. Minimum Quantifiable Limit (MQL) is defined as that concentration of analyte sufficiently in excess of MDL such that a numerical value can be assigned to the analyte. MDL and MQL are theoretical values which we normally have based on a blank value exhibited by the on-line monitor. When a blank value is observed, MDL and MQL are established as 3 and 10 times the standard deviation of the blank value. However, for the IRT chloroprene prestart study a zero value was obtained for blank measurements. In the absence of a finite blank value the lowest concentration of chloroprene that could be practically maintained in an exposure chamber was used to determine MDL and MQL. This was accomplished as described in the section on determination of MLQ.

The Minimum Limit of Quantitation (MLQ) is defined as the lowest concentration of analyte that can reliably be assigned a numerical value with an RSD of $\leq 10\%$ and a relative error of $\leq 10\%$. Therefore, to determine MLQ, grab sampling must be performed to allow a comparison between the on-line monitor determination and a reference method assumed to yield the true value for chloroprene concentration.

The Quantitation Limit (QL) is defined as the concentration below which acceptable quantitation cannot be achieved. The quantitation limit is usually defined the same as the MLQ. However, in some cases where MLQ is near the lowest exposure concentration, the QL may be assigned the same value as MQL. Measured values below the QL are not normally reported in daily summaries.

2. Determination of MDL and MQL

Analysis of the charcoal filtered air blank by the on line monitor while the stream select valve was in normal rotation failed to detect chloroprene. To determine MDL and MQL, the precision measurements were performed by monitoring a chamber with a low chloroprene concentration. A standard deviation of 188 area counts was found for a chloroprene concentration measured at 0.88 ppm by the on-line monitor (0.73 ppm by grab sampling). MDL and MQL are set at three and ten times the standard deviation expressed in concentration units. On this basis MDL and MQL were 0.06 and 0.20 ppm respectively.

3. Determination of MLQ

a. Experimental

Sampling to determine MLQ was done by taking grab samples with DMF filled bubblers. The bubbler samples were analyzed on an off-line GC calibrated using gravimetrically prepared chloroprene standards. Each standard and

bubbler had a constant amount of internal standard added. The purpose of the internal standard was to compensate for any fluctuations in sample volume and injection volume. Grab samples were taken from the low concentration chamber. The vapor delivery rate into the exposure chamber was reduced so that concentrations of about 10, 5, 2 and 1 ppm could be achieved.

Gravimetric standards were prepared so that the standards bracketed the expected concentration of chloroprene to be sampled at each level. The standards and samples were analyzed by GC using a procedure similar to that used to analyze grab samples acquired from the chamber for purposes of on-line monitor calibration.

b. Results

The off-line GC showed good sensitivity and precision for the low gravimetric standard. The relative response factor for the standards was used to determine the amount of chloroprene collected in the bubblers. Based upon the criteria that MLQ is the concentration of analyte for which the relative standard deviation is $\leq 10\%$ and the relative error is also $\leq 10\%$, MLQ was set to 5 ppm using the data shown in Table A.1. The established MLQ was 50% of the low exposure chamber concentration.

4. Determination of QL

Since the established MLQ of 5 ppm was 50% of the low exposure chamber concentration, the QL was set to 0.2 ppm which was the MQL.

E. Precision, Linearity and Absolute Recovery of the On-Line Monitor

Precision of the on-line monitor was estimated from measurements of the on-line standard sampled from each of the 12 sample valve ports. These measurements had a relative standard deviation of 0.5% (N=12). In addition, in order to estimate precision for the on-line monitor at each concentration level, repeated measurements were made from each exposure chamber (at the original target concentrations). The following values were obtained:

<u>Location</u>	<u>Number of Measurements (N)</u>	<u>Mean (ppm)</u>	<u>Standard Deviation (SD)</u>	<u>Relative Standard Deviation (RSD)</u>
125 ppm chamber	5	117.19	0.14	0.12%
125 ppm chamber	5	122.73	0.11	0.09%
25 ppm chamber	5	23.03	0.09	0.40%
25 ppm chamber	5	24.61	0.09	0.38%
5 ppm chamber	5	4.81	0.01	0.27%
5 ppm chamber	5	5.00	0.01	0.16%

TABLE A.1. Chloroprene Inhalation Developmental Toxicity Study: Chloroprene
MLQ Determination Results.

Grab Conc. ppm	On-Line Monitor Conc (ppm)	Error (ppm)	Relative Error	Monitor Response	Monitor (%RSD)
10.33	10.08	0.25	2.4%	96,411	1.1
5.61	5.19	0.42	7.5%	49,598	0.5
2.43	1.88	0.55	22.6%	18,075	0.4
1.21	0.86	0.35	28.9%	8,390	2.2

The precision results reported above for the measurements made on the chamber concentrations were affected by both variations in test material concentration in the chamber and instrument imprecision of the on-line monitor. However, the overall precision was adequate to demonstrate adequate instrument precision at each sampling point. The excellent precision seen for the on-line standard also indicated good reproducibility for the on-line monitor. Performance of these tests at the initial target concentrations does not compromise this validation of instrument precision. That is, instrumental precision should not change due to the difference in concentration between the initial and revised dose levels.

Linearity of the on-line monitor was determined by calibrating the on-line monitor against a gravimetrically calibrated GC (see "Calibration of the On-Line Monitor" section). The on-line monitor calibration was linear through the range of interest.

Absolute recovery of the bubbler grab sampling method was confirmed by obtaining grab samples with backup bubblers from each of the exposure chambers. An average breakthrough of less than 5% was detected. The determination of chamber concentration by analysis of grab samples was corrected to account for breakthrough.

F. Monitoring for Chloroprene in Building Exhaust

Effluent from the exposure chambers and the exposure generation system was diluted with the exhaust air of the entire LSL-II building prior to exhausting from the building stack. The expected concentration of chloroprene in the building exhaust was less than 0.11 ppm. The chloroprene concentration in the exhaust of the building was determined using grab sampling employing standard charcoal gas sampling tubes and desorbed with DMF. Good recovery and trapping of chloroprene using charcoal tubes was shown during the chemical stability studies. Three samples were collected from different locations within the exhaust stream. The average building exhaust concentration of chloroprene was shown to be 0.09 ppm with a SD of 0.01 ppm for the three samples.

Test Chemical Stability in the Exposure System

III. TEST CHEMICAL STABILITY IN THE EXPOSURE SYSTEM

A. Introduction

In exposure systems designed to generate organic test chemical vapors, BNW frequently employs a common approach to the identification and analysis of potential impurities in the exposure atmosphere. First, the generation system can cause degradation of the test chemical when high temperatures must be employed to vaporize the required amount of test chemical into the exposure system. This can result in thermal decomposition or polymerization of the test chemical. Moreover, the products of such thermal decomposition or polymerization may also be subject to further thermal degradation forming other volatile components. Secondly, volatile impurities in the test chemical are evaluated as possible contaminants of the generated test atmosphere. This source of contamination can be especially significant when a test chemical impurity exhibits a vapor pressure which is substantially higher than that of the test chemical. Relative to the test material, concentrations of highly volatile impurities can become enriched in the exposure atmosphere, even when present in the original test chemical at low concentrations.

Chloroprene is quite reactive and known to polymerize in the vapor generator. The test material was fortified with inhibitors to limit polymerization and reduce peroxide formation. However, the generator design provided for removal of less volatile inhibitors and degradation products. Thus, the details of the generator design and operation are a critical ingredient to the discussion of test material stability and the composition of the atmosphere which was presented to the animals. Generator design is discussed elsewhere, however in brief: achievement of exposure levels was obtained by passing a nitrogen stream over a pool of heated chloroprene. The nitrogen stream exited the generator through a refluxing condenser and was directed down a distribution line. This refluxing action removed impurities and degradation products less volatile than chloroprene from the generator effluent. A portion of the distribution line flow was directed to each chamber to provide the required concentration.

Chloroprene undergoes both dimerization and rapid reaction with oxygen to form peroxides. Chloroprene peroxides serve as a free radical source and can initiate runaway exothermic polymerization. Peroxide formation is reduced by the use of tert-butylcatechol (TBC), which acts as an oxygen scavenger. Polymerization, once initiated, is limited by inhibition with phenothiazine, a free radical scavenger. The chloroprene used in this study was inhibited at the 0.1% level with both TBC and phenothiazine. Maintenance of a nitrogen blanket and the use of inhibitors keep the formation of peroxides and polymer to a minimum. These measures were effective in preventing polymer formation during the repeated dose and subchronic studies performed earlier at BNW.

However, despite the ability of the measures outlined above to reduce peroxide and polymer formation, it is important to recognize that the rate of dimer formation is dependent upon temperature and is not influenced by the presence of inhibitors. It is reported that the rate of dimer formation is about 0.35% per day at room temperature and about 1% per day at the boiling point of chloroprene (Kirk-Othmer, 1979, Encyclopedia of Chemical Technology, 3rd Edition, Vol 5). Thus, substantial amounts of dimer were formed in the vapor generator evaporation flask.

The literature (Stewart, CA, 1971, J. Amer. Chem. Soc., 93: 4815-4821) reports the elution pattern for a total of six dimer isomers on a SE-30

column, two of the dimer isomers co-eluted ((1-chloro-4-(1-chlorovinyl)cyclohexene and 2-chloro-4-(1-chlorovinyl)cyclohexene). The SE-30 column employed by Stewart is a dimethyl silicon stationary phase of relatively low polarity (2 ft SE-30, 20% on Diaport S support). An injection port temperature of 150°C and a FID temperature of 175°C were used to "minimize thermal destruction of sensitive compounds." At both ~20°C and at 60°C more trans-1,2-dichloro-1,2-divinylcyclobutane was formed than any other dimer isomers (from 30 to 42% of the total dimer content, depending upon temperature and extent of conversion with lower amounts formed at 60°C). The dimer isomer formed in the greatest quantity demonstrated the shortest chromatographic retention time. Stewart also concluded that the six isomers reported represent the predominate products formed at room temperature dimerization, "it is believed that no other products are present in room temperature dimerization in an amount greater than 1% of the mixture." Three of the dimer isomers are present in about equal amounts. The remaining divinylcyclobutane adduct (cis-1,2-dichloro-4-vinylcyclobutane) readily undergoes rearrangement to form 1,6-dichloro-1,5-cyclooctadiene; none of this rearrangement product is formed directly by initial monomer dimerization. At higher temperatures the rearrangement and dechlorination of other dimers becomes significant with formation of dehydrochlorination products.

In the repeated dose and subchronic studies BNW found that dimer concentration in the chambers was about 0.01% of the chloroprene concentration, even though the concentration of dimer rose to about 6% in the evaporation flask by the end of the day. This is a result of the refluxing nature of the vapor generator and the wide differences in boiling point between chloroprene and the various dimer isomers, i.e. fractionation in the generator purifies the chloroprene vapor.

A significant increase in the dimer concentration in the distribution line was not expected to occur unless the condenser malfunctioned (resulting in loss of separating capability), or if the contents of the generator flask became quite depleted in chloroprene and significantly enriched in dimer. Taken to the extreme, once all of the monomer is "distilled over" then the less volatile dimer isomers will exhibit greater partial pressures. Thus, it was important to run the generator so that an adequate "heel" of chloroprene remained at the end of each generation day. BNW's daily operation procedure for the generator was designed to ensure that dimer concentration was not excessively enriched at the end of a particular exposure day.

Two measures were implemented to minimize chamber dimer concentrations. First the generator was charged with enough chloroprene to ensure that about 150 grams remained at the end of the exposure day. If the start of the exposure was excessively delayed, the generator was shut down and additional chloroprene added to the flask. The other measure which ensured that a low animal exposure to dimer was maintained was the use of the dedicated on-line monitor to determine test material purity throughout each exposure day. BNW implemented a second on-line gas chromatographic monitor to repeatedly sample the distribution line throughout the exposure day. A longer GC run time was required than used on the on-line chamber monitoring GC in order that both very volatile (1,3-butadiene and 3-buten-1-yne) and less volatile isomers (such as dimer) could both be resolved. In the event that some inadvertent change in the operation of the generator increased the dimer content of the generator output, then the on-line purity determination allowed the exposure personnel to shut down the vapor generator.

In summary, the majority of the impurities expected in the test chemical exhibit boiling points greater than chloroprene, and were thus not expected to

occur in measurable quantities in the exposure atmospheres. Of the known impurities in chloroprene, only 1,3-butadiene and 3-buten-1-yne were subject to enrichment from selective vaporization in the generated exposure atmospheres. These impurities were reduced to less than 0.5% of the generated vapor before animal exposure was allowed to proceed. Most of the volatile products of thermal decomposition are also less volatile than chloroprene and were expected to remain in the generator reservoir. Impurities present in the bulk test material that are similar in volatility to chloroprene and could not be removed by this method of generation include chlorobutene and cis/trans 1-chlorobutadiene.

Studies of the chemical stability of chloroprene in the generator reservoir, distribution line and exposure chambers were conducted in both empty and occupied chambers. The chamber samples were collected within the first and last hour of the generation day. The characterization of chemical stability included use of gas chromatography mass spectrometry (GC/MS) to identify dimer and other organic impurities in an aged sample of test material and gas chromatography to measure the amounts of impurities present in the chambers, distribution line and generator flask samples obtained at the beginning and end of the day. During the prestart, peroxide content was determined in test material from the generation flask at the beginning and end of the exposure day and measurement of inhibitors in samples acquired from the distribution line was accomplished by gas chromatography. In addition, an on-line GC method was implemented to measure the purity of the chloroprene vapor in the distribution line.

B. Experimental Methodology and Results

1. Determination of Chloroprene Purity in the Generation Flask.

For exposure generation, a one liter evaporation flask was filled (under a nitrogen blanket) once at the beginning of each exposure day with ~530 grams of chloroprene from one of the original 4-liter glass containers. Stability of chloroprene in the generator generation flask was demonstrated by determining the purity of chloroprene in the generation flask over the time period that the material was contained in the generation flask during normal study operation.

Purity analysis and peroxide content of the generation flask samples was accomplished using the same methods used for the routine bulk chemical reanalysis of chloroprene. A chloroprene sample was removed from the generation flask immediately after filling at the beginning of the day. An additional chloroprene sample was collected at the end of the exposure period.

The purity assay of samples from the generation flask was performed on a HP 5890 GC equipped with a FID and a DB-5 capillary column, 30 m x 0.53 mm i.d. fused silica, 1.5 μ film thickness. The peroxide content of the generation flask samples was determined by potentiometric titration of samples with standardized sodium thiosulfate. Complete instrument parameters can be found in SOP 0B-AC-3A14 "Bulk Analysis of Chloroprene".

The chloroprene in the generation flask immediately after filling was determined to be 98.5% pure by area percent. The area percent purity of the material at the end of generation on the same day was determined to be 90.6%, the most concentrated dimer isomer was present at about 2% while the total dimer content was about 5-6%. The peroxide content in the sample collected

immediately after filling was 0.05 meq/kg. The peroxide content in the sample collected at the end of generation on the same day was 0.14 meq/kg.

2. Determination of 4-TBC and Phenothiazine in the Distribution Line

The chloroprene exposure material contained ~1,000 ppm 4-tertbutylcatechol (4-TBC) as an anti-oxidant and ~1,000 ppm phenothiazine as an inhibitor to polymer formation. The design of the generation system and the fact that 4-TBC and phenothiazine are much less volatile than chloroprene ensured that 4-TBC and phenothiazine were not present at significant levels in the exposure atmosphere. Efficient removal of 4-TBC and phenothiazine from the vapor generator output was confirmed by GC analysis of samples from the distribution line. Ten liter samples (containing about 3.3 g of chloroprene) were collected from the exposure distribution line in DMF filled bubblers within the first hour and the last hour of generation on a normal six hour generation day (samples acquired 3/15/90; BNW NB 53436 pg 92).

Recovery of 4-TBC and phenothiazine was confirmed by spiking a bubbler with a known amount of 4-TBC and phenothiazine and pulling 10 liters of room air through the bubbler. The recovery study indicated no loss of 4-TBC or phenothiazine for a 10 liter air sample. The relative amount of inhibitor and detection limits were calculated relative to the sample size of chloroprene. The amount of chloroprene collected was calculated from the sample volume and the concentration of chloroprene present in the distribution line.

The samples were analyzed relative to gravimetrically prepared 4-TBC and phenothiazine standard solutions using a HP 5890 gas chromatograph with a FID and a 30 meter x 0.53 μ meter, 1.0 μ meter film, DB-5 capillary column (J&W). Azobenzene was used as an internal standard. The detector was operated at 275°C. One microliter cool on-column injections were made with an initial oven temperature of 130°C. The oven temperature was ramped to 250°C at 15°C/minute. See Table A.2 for complete GC conditions. A linear calibration curve was observed for both sets of standards.

No 4-TBC or phenothiazine were detected in these samples, the amounts were below that of the lowest concentration standard which produced a detectable response. Using the concentration of this standard (equivalent to 200 μ g in a bubbler sample) as a detection limit, the amount of 4-TBC and phenothiazine in the distribution line was shown to be less than 0.006% (w/w) with respect to chloroprene. This equates to a distribution line concentration of ~3 ppm 4-TBC and phenothiazine. Allowing for the dilution of the distribution line to the exposure chambers, the concentration of 4-TBC and phenothiazine in the 175 ppm chamber, 40 ppm chamber and the 10 ppm chamber would be less than 120 ppb, 30 ppb and 6 ppb, respectively.

3. Determination of Trace Contaminants or Degradation Products in the Exposure Atmosphere and the Generation Reservoir

The possible presence of trace amounts of contaminants or degradation products was investigated in samples collected from various points in the exposure generation system. Analysis was by gas chromatography. Samples were collected with standard charcoal gas sampling tubes from the 10 and 175 ppm exposure chambers and the distribution line during the first and last hours of generation on a normal six hour generation day during the study. The charcoal tubes were desorbed with dimethylformamide. Standards of suspected degradation product and impurities were not available.

TABLE A.2. Chloroprene Inhalation Developmental Toxicity Study: Gas Chromatographic Conditions Used for the Analysis of Chloroprene Distribution Line Samples on 3/29/90.

Gas Chromatograph:	Hewlett Packard 5890A
Column:	30 meter x 0.53 mm I.D. Megabore DB-5 1.0 μ meter film (J & W Scientific)
Initial Temperature:	130°C
Initial Time:	5.00 minutes
Rate:	15°C/minute
Final Temperature:	250°C
Final Time:	5:00 minutes
Detector Type:	Flame Ionization
Detector Temperature:	275°C
Injector:	On-Column
Injection Volume:	1 μ l

Sample size was approximately 0.25 liters for the distribution line, 7 liters for 175 ppm chamber and 10 liters for the 10 ppm chamber. Breakthrough was found to be less than 1% for all charcoal samples by analysis of the secondary charcoal bed within the tubes. We assume that good trapping efficiency for isomers such as dimer will be achieved when good trapping efficiency is observed for chloroprene. Comparison of a chloroprene standard with a chloroprene standard desorbed from charcoal shows a greater than 90% average recovery ratio from the charcoal.

The samples were analyzed using a HP 5890 gas chromatograph with a FID and a 30 meter x 0.53 μ meter, 1.5 μ meter film, DB-5 capillary column (J&W). The detector was operated at 300°C and the injector was operated at 250°C. Duplicate injections were made using a HP 7673A autosampler.

Assignment of dimer and inhibitor peaks was verified by GC/MS. The complete GC/MS report is located in this appendix. Compounds identified as chlorobutene and 1-chlorobutadiene were detected in all samples at approximately 0.1 to 0.8% by area. No dimers or inhibitors were present in the distribution or chamber samples at greater than 0.01% of the total sample area. An unidentified compound was noted in the samples collected during the first hour of generation from the distribution line and chamber samples on 3/29/90 that was not observed in samples collected the last hour of the generation period from the chambers or distribution line. This compound exhibited a retention time of ~15.6 minutes compared to a retention time of ~4.6 minutes for chloroprene. In the samples acquired at the start of the generation day this compound was found to be present at between 0.002 and 0.13% by area relative to chloroprene. No additional compounds were noted with area response amounts of greater than 0.11% relative to chloroprene (percent of total peak area). Results of the analysis of distribution line and chamber samples are summarized in Table A.3.

4. On-Line Determination of Chloroprene Purity

The purity of chloroprene vapor directed to the exposure chambers was monitored every exposure day using a separate HP5890 gas chromatograph (on-line purity GC). The on-line purity GC sampled an ~10,000 ppm stream of chloroprene produced by an air dilution of the ~90,000 ppm generated chloroprene vapor. By sampling this high concentration stream of chloroprene relatively small amounts of impurities were easily detected.

The on-line purity GC was used to insure that volatile compounds were vented to waste before initiation of animal exposures. Once exposures were initiated the GC parameters were changed to allow both volatile and less volatile impurities (including dimers) to be determined. This allowed daily confirmation that the study animals were not subject to significant concentration of chloroprene dimer or the more volatile impurities found in chloroprene.

The daily operation of the purity GC was controlled by a BASIC program running in the background. At the start of the exposure day the program loaded and executed a short/low temperature analysis of volatile compounds in the generated chloroprene. When the area percentage of the major volatile peak was less than 0.5% and the area percent of the chloroprene was greater than 98.5% the BASIC program ramped the GC oven temperature to clear trapped compounds and began temperature programmed analyses of the chloroprene vapor to allow monitoring of both volatile compounds and less volatile compounds including the dimers of chloroprene. Finally, after the termination of generation the BASIC program directed the purity GC to monitor a sample

TABLE A.3. Chloroprene Inhalation Developmental Toxicity Study: Composition of Degradation Samples by GC Area %.

Sample	Time	Chloroprene	Chlorobutene	1-Chlorobutadiene
Distribution Line (3/29/90)	a.m.	99.3%	0.5%	0.04%
175 ppm Chamber (3/29/90)	a.m.	99.2%	0.6%	0.05%
10 ppm Chamber (3/29/90)	a.m.	99.2%	0.5%	0.03%
Distribution Line (3/29/90)	p.m.	99.1%	0.7%	0.08
175 ppm Chamber (3/29/90)	p.m.	99.1%	0.8%	0.1%
10 ppm Chamber (3/29/90)	p.m.	99.1%	0.8%	0.04%

Note: No dimers or inhibitors were present in the distribution or chamber samples at greater than 0.02% of the total sample area.

containing a low amount of dimers and a room sample for a blank. The instrument parameters for the on-line purity GC are shown in Table A.4.

A diffusion generator, used to produce a chloroprene dimer vapor, was set up using material from the chloroprene generator at the end of a test generation day (BNW 12377-45). Removal of the majority of the chloroprene from the dimers was achieved using nitrogen sparging. The diffusion generator was loaded with a single cell containing the high dimer material and operated at a diffusion cell temperature of 50°C and a total flow of 183 ml/min of nitrogen (BNW 53436-96). This source of dimer vapor allowed confirmation of the retention time of the predominate dimer isomers and an estimation of the detection limit for dimer by the on-line purity method.

A chromatogram from the start of the test exposure day (short/low temperature analysis) is given in Figure A.3 (top chromatogram). Chloroprene was found to elute at about 2 minutes on this system. The on-line purity GC allowed the separation and area percent quantification of 5 compounds more volatile than chloroprene. The total area percent of these compounds was found to be less than 0.65% at the start of the day and less than 0.04% at the end of the day. A large percentage (~60%) of the volatile compounds was due to a single peak eluting at 0.9 minutes believed to be 1,3-butadiene or 3-buten-1-yne or a mixture of the two compounds. Two compounds less volatile than chloroprene were detected. These compounds (believed to be 1-chlorobutadiene and isomers of chlorobutene) were present at about 0.7% at the start of exposure and 0.5% (by area percent) at the end of exposure.

A chromatogram from the end of the test exposure day using temperature programmed analysis is shown in the middle chromatogram in Figure A.3. No chloroprene dimer peaks were observed. The small peaks seen at the end of the run were background peaks and were seen in the analysis of blank samples.

A chromatogram obtained from the on-line purity GC by sampling the dimer diffusion generator output is shown in the bottom chromatogram of Figure A.3. The peak at 1.429 minutes is methylene chloride which was used to transfer the dimers to the diffusion generator. Three dimer peaks eluted at about 9.9, 11.3 and 12.3 minutes. These retention times agree with retention times of the three most abundant chloroprene dimers. Retention time mapping of the dimers was achieved by the comparison of the direct injection of a very small amount of neat chloroprene into the on-line purity GC (3/13/90 run #298) with GC/MS analysis on a similar column (the complete GC/MS report is contained in the Appendix). The large to small graded peak area of the dimers on the on-line purity GC was due to the enhancement of more volatile compounds by the diffusion generator.

During the prestart, the average area percent purity determined by the on-line GC for chloroprene vapor generated on 3/15/90 (a typical test exposure day) was $99.18 \pm 0.34\%$. The on-line purity GC showed an average % purity of 99.34% for the study. An area percent purity determination is valid when the eluted compounds show the same flame ionization detector (FID) response as chloroprene. The FID response of the compounds found in chloroprene is similar to chloroprene since they are structurally similar chloroprene in the amount of oxidation, unsaturation, and halogenation. Dimer was detected at the end of only three exposure days during the study and was present at less than 0.1%. The purity GC was estimated to have a detection level for dimers of ~0.04% of the chloroprene peak area. This indicated that the chloroprene vapor used for animal exposures had an average purity greater than 99% and generally contained less than 0.04% (w/w) dimer, thus the 175 ppm chamber contained less than 0.035 ppm (v/v) of the predominate dimer isomer.

TABLE A.4. Chloroprene Inhalation Developmental Toxicity Study: Instrument Parameters for On-line Determination of Chloroprene Vapor Purity.

Equipment and Equipment Parameters

Gas Chromatograph	HP5890
Detector	FID
Integrator	HP3393
Detector Temperature	300°C
Sample Valve	no purge 1/16" Hastelloy C valve equipped with a 0.5 ml nickel loop
Stream Select	12 port 1/16" Hastelloy C valve
Valve Box Temperature	100°C
Column	30 meter DB-5 megabore (0.53 µm ID) fused silica capillary column with 1.5 µm film BNW 12379-104 S/N 8194724
Carrier	nitrogen (oxygen and hydrocarbon filtered)
Carrier Flow	~10.2 ml/min
Column Head Pressure	4 psi

Short Runtime Analyses Parameters

Initial Oven Temperature	30°C
Initial Time	3 min
Run Length	3 min

Long Runtime Analyses Parameters

Initial Oven Temperature	30°C
Initial Time	3 min
Temperature Ramp	15°C/min
Final Temperature	275°C
Final Time	5 min
Run Length	24.33 min (w/o cooling time required to return to the initial oven temperature - the total cycle time is ~40 min)

C. Discussion

Gas chromatography showed that the purity of the test material in the generation flask at the end of the generation day was substantially lower than the purity at the beginning of the day. This was due to formation and concentration of dimer isomers in the generation flask.

One impurity peak at less than 0.13% (as percent of total peak area) was observed in charcoal tube samples acquired from the chambers and distribution line during the first hour of the exposure day. The unknown eluted after chloroprene and before the dimer peaks. This unknown was not observed with the on-line purity method. We do not believe this isolated observation indicated significant exposure of the test animals to this unknown product.

Trace amounts of chlorobutene and 1-chlorobutadiene were found in samples collected from the chambers and the distribution line. The amounts of these impurities were found to be less than 0.8% by area. The inhibitors, 4-TBC and phenothiazine were present in the distribution line at approximately 3 ppm. Dimers were not detected in the distribution or chamber samples by either the analysis of charcoal tube samples or the on-line purity method. The detection limits of dimer were estimated to be ~0.02% by the charcoal tube method and ~0.04% by the on-line method (as percent of the total peak area of the most abundant dimer isomer). Test chemical stability and purity were considered acceptable for the study.

GC/MS Analysis of Chloroprene and Its Dimer Products

DATE March 13, 1990

TO Barry Hayden

FROM RB Lucke *RB Lucke*

SUBJECT GC/MS analysis of chloroprene and its dimer products..

One samples, BNW 53436-72-2 was analyzed by GC/MS. All pertinent information was recorded in BNW# 12410 pages 39-42. Hardcopy data is stored in the GC/MS file in room 2 of 329 building while the electronic-data is archived on magnetic tape in Lab 15a of the 329 building.

Instrumentation:

- * Hewlett Packard 5988 GC/MS.
- * Hewlett Packard 5890 GC.
- * Hewlett Packard RTE-6 data station.
- * GC conditions: The column used was a DB-1 capillary GC column, 30 meter by 0.25 mm i.d. by 1.0 μ m film thickness (J&W serial # 81665). Injection was in the splitless mode at 200° C, 0.6 min purge. The oven profile started at 35° C for 5 minutes followed by a 10° C per minute ramp to 275° C with a final hold time of 5 min.
- * Mass Spectrometer Conditions: Operated in the Electron Impact mode (EI) at 70eV. The mass range was scanned from 40 - 400 amu with the electron multiplier set at 2643 volts (300 volts above autotune) and a 300 μ A emission current. The source temperature was set at 200° C and the interface was at 200° C. The GC/MS was autotuned with PFTBA prior to analysis for instrument calibration. In addition, 1.2 ng of methyl stearate was injected into the GC/MS to verify instrument performance. Mass spectra identification was made by comparison of spectra from individual standards, the EPA/NIH library and/or from conventional mass spectral interpretation.

DATA and RESULTS:

This includes 17 figures attached to this letter report.

[Referred to as FIGURES A.4.1 through A.4.17]

The GC/MS total ion chromatogram compares very well with the GC/FID chromatogram (Figures 1 and 2). Figures 5 through 12 show the mass spectra and the library hits. Mass spectral comparison with that from the EPA/NIH library indicates that all the peaks are chloroprene dimers with the exception of *terbutylcatechol* and *phenothiazine*. The most abundant ion in most all of the mass spectrum is the 88 atomic mass unit (amu) ion. Figure 16 shows the extracted ion chromatogram for 88 amu and compares very well with the TIC in figure 15. This indicates that the peaks are of a similar molecule that fragment in a similar way.

Analysis Cost:

Work order number M38969 was charged 7 units of GC/MS usage (\$214 / unit). This included instrument and operator time spent directly on the project.

53436-72-2 O.Sul nest injection

RUN# 94

MAR 11, 1998 16:22:23

"Aged" Chloroprene

AREAX

RT	AREA	TYPE	WIDTH	AREAX
1.695	21633	PV	.023	.00352
1.741	49733	VV	.026	.01278
1.775	138164	VV	.028	.03527
2.189	64389	VB	.035	.01644
2.347	38428	VB	.035	.00981
3.195	24827792	PV	.041	6.13446
3.372	368119848	VB	.097	91.94186
3.476	1788677	BB	.028	.43419
3.727	255815	BB	.065	.06511
11.465	6889	BB	.061	.00133
17.166	1283335	PB	.056	.32764
17.628	8924	BP	.059	.00228
17.981	85278	PB	.056	.02177
19.858	59587	BV	.058	.01519
19.282	841563	VB	.057	.21486
19.838	868841	PV	.064	.22162
19.897	754223	VB	.056	.19256
20.228	9335	BV	.067	.00238
21.755	21168	PV	.052	.00548
21.857	576625	VB	.062	.14722
24.877	333623	PB	.064	.08518
30.148	1374	BB	.043	.00035
32.135	421485	PB	.104	.18761

FIGURE A.4.1. Chloroprene Inhalation Developmental Toxicity Study: GC/FID Chromatogram Provided by B.K. Hayden.

FIGURE A.4.2. Chloroprene Inhalation Developmental Toxicity Study: GC/MS
Total Ion Chromatogram of Sample 53436-72-2.

FIGURE A.4.3. Chloroprene Inhalation Developmental Toxicity Study: GC/MS TIC of Sample 53436-72-2 With Peak Numbers.

MS data file header from : >L3122

Sample: Operator: RICH MS 3/12/90 15:53
 Misc : BTL# 1
 Sys. #: 1 MS model: 88 SW/HW rev.: 1A ALS #: 0
 Method file: BARRY1 Tuning file: APE101 No. of extra records: 2
 Source temp.: 200 Analyzer temp.: 250 Transfer line temp.: 0

Chromatographic temperatures : 35. 275. 0. 0. 0.
 Chromatographic times, min.: 5.0 10.0 0.0 0.0 0.0
 Chromatographic rate, deg/min: 10.0 0.0 0.0 1.0 0.0

>L3122

40.01 400.0 ADC TIC
 Upslope: .20 Area Reject: 5.00 % Max Peaks: 8 Bunching: 1
 Dnslope: 0.00 Results File ILB014 Sorted by Time/Area INT

Peak #	R.T. min.	first scan	max scan	last scan	peak height	raw area	corr. area	corr. % max.	% of total
1	17.59	844	863	865	309440	3067935	3016933	86.09	26.81
2	18.12	888	897	899	53397	371597	219843	6.27	1.95
3	19.36	961	975	977	269867	1963426	1939393	55.34	17.23
4	19.89	992	1009	1011	386963	3525952	3504523	100.00	31.14
5	19.96	1011	1013	1016	369245	757139	734583	20.96	6.52
6	21.52	1099	1112	1115	183653	1064401	1039985	29.68	9.24
7	23.52	1228	1239	1242	71876	328776	328776	9.38	2.92
8	30.56	1669	1685	1689	87264	467243	467243	13.33	4.15

Sum of corrected areas: 11251282.

5:15 PM MON., 12 MAR., 1990

FIGURE A.4.4. Chloroprene Inhalation Developmental Toxicity Study:
 Integration Report Table of Figure A.4.3.

1. Benzenemethanamine, 2-chloro- (9CI)	141 C7H8ClN
2. 1,3-Butadiene, 2-chloro- (8CI9CI)	88 C4H5Cl
3. 3,5-Heptadiyn-2-one (8CI9CI)	106 C7H6O

Sample file: >L3122 Spectrum #: 863
Search speed: 2 Tilting option: S No. of ion ranges searched: 57

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_
1.	20*	89974	15818	"B1808	42	70	2	1	72	55	5	:
2.	20*	126998	6992	"B1808	36	79	2	0	100	55	5	:
3.	15*	13879715	2199	"B1808	39	46	2	1	29	58	3	:

FIGURE A.4.5. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #1.

1. 1,3-Butadiene, 2-chloro- (8C19C1) 88 C4H5Cl
2. 1,5-Cyclooctadiene, 1,6-dichloro- (8C19C1) 176 C8H10Cl2

Sample file: >L3122 Spectrum #: 897
Search speed: 2 Tilting option: S No. of ion ranges searched: 55

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	42*	126998	6992	"BIGDB	35	80	1	0	81	30	14	1
2.	25*	29480420	7092	"BIGDB	35	80	3	0	100	49	7	1

FIGURE A.4.6. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #2.

1.	1,3-Butadiene, 2-chloro-, dimer (8C19CI)	0	
2.	Cyclohexene, 1-chloro-4-(1-chloroethenyl)- (9CI)	176	CBH10CI2
3.	1,5-Cyclooctadiene, 1,6-dichloro- (8C19CI)	176	CBH10CI2
4.	Benzenamine, 3-chloro-2-methyl- (9CI)	141	C7H8CIN
5.	1,3-Butadiene, 2-chloro- (8C19CI)	88	C4H5Cl

Sample file: >L3122 Spectrum #: 975
Search speed: 2 Tilting option: S No. of ion ranges searched: 55

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	67	14523898	7174	"B1GDB	86	55	2	0	66	11	34	2
2.	57*	13547063	7173	"B1GDB	44	74	0	2	58	35	22	2
3.	47*	29480420	7092	"B1GDB	35	80	0	0	61	32	20	2
4.	15*	87605	15957	"B1GDB	43	58	2	1	34	60	3	1
5.	15*	126998	6992	"B1GDB	35	80	2	0	79	60	3	1

FIGURE A.4.7. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #3.

1.	Cyclohexene, 1-chloro-5-(1-chloroethenyl)- (9CI)	176	CBH10C12
2.	1,3-Butadiene, 2-chloro-, dimer (8CI9CI)	0	
3.	2-Butene, 1-isothiocyanato- (9CI)	113	C5H7NS
4.	Cyclohexene, 1-chloro-4-(1-chloroethenyl)- (9CI)	176	CBH10C12
5.	1,5-Cyclooctadiene, 1,6-dichloro- (8CI9CI)	176	CBH10C12

Sample file: >L3122 Spectrum #: 1009
Search speed: 2 Tilting option: S No. of ion ranges searched: 55

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	72*	13547074	11235	"B1808	72	62	0	2	62	31	32	7
2.	47*	14523898	7174	"B1808	68	57	2	2	43	43	16	4
3.	26*	2253932	11039	"B1808	24	68	3	0	323	40	10	1
4.	23*	13547063	7173	"B1808	43	59	0	2	43	60	6	4
5.	21*	29480420	7092	"B1808	40	59	0	2	42	60	5	3

FIGURE A.4.8. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #4.

1. Cyclohexene, 1-chloro-4-(1-chloroethenyl)- (9CI) 176 C8H10Cl2
2. 1,3-Butadiene, 2-chloro-, dimer (8CI9CI) 0
3. 1,5-Cyclooctadiene, 1,6-dichloro- (8CI9CI) 176 C8H10Cl2
4. 1,3-Butadiene, 2-chloro- (8CI9CI) 88 C4H5Cl
5. 2-Butenoic acid, 4-oxo-4-phenyl- (9CI) 176 C10H8O3

Sample file: >L3122 Spectrum #: 1013
Search speed: 2 Tilting option: S No. of ion ranges searched: 55

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_1	R_1U
1.	94*	13547063	7173	"B1GDB	87	31	0	0	81	13	64	95
2.	79*	14523898	7174	"B1GDB	81	60	2	0	86	13	43	61
3.	73*	29480420	7092	"B1GDB	54	61	0	0	69	23	32	66
4.	12*	126998	6992	"B1GDB	41	74	1	0	58	62	2	21
5.	12*	583062	21059	"B1GDB	49	49	2	1	68	65	2	21

FIGURE A.4.9. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #5.

1. 1,5-Cyclooctadiene, 1,6-dichloro- (8C19Cl)	176 C8H10Cl2
2. 1,3-Butadiene, 2-chloro- (8C19Cl)	88 C4H5Cl
3. 3,5-Heptadiyn-2-one (8C19Cl)	106 C7H6O

Sample file: >L3122 Spectrum #: 1112
Search speed: 2 Tilting option: S No. of ion ranges searched: 61

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	58	29480420	7092	"B16DB	60	55	0	0	100	17	25	2
2.	23*	126998	6992	"B16DB	42	73	0	0	58	58	6	4
3.	15*	13879715	2199	"B16DB	38	47	2	1	26	57	3	1

FIGURE A-4-10. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #6.

1. 1,2-Benzenediol, (1,1-dimethylethyl)- (9CI)	166 C10H14O2
2. 1,2-Benzenediol, 4-(1,1-dimethylethyl)- (9CI)	166 C10H14O2
3. Silane, trimethylphenoxy- (8CI9CI)	166 C9H14OSi
4. Ethanone, 1-(2-hydroxy-6-methoxyphenyl)- (9CI)	166 C9H10O3
5. Benzaldehyde, 4-methoxy-, oxime (9CI)	151 C8H9NO2

Sample file: >L3122 Spectrum #: 1239
Search speed: 2 Tilting option: S No. of ion ranges searched: 55

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	94*	27213781	17502	"B1GDB	83	11	0	1	79	8	68	9
2.	52*	98293	17495	"B1GDB	42	54	2	0	91	16	20	1
3.	52*	1529175	17499	"B1GDB	45	43	2	0	69	23	22	2
4.	37*	703231	17488	"B1GDB	37	69	2	0	76	26	14	1
5.	33*	3235049	17454	"B1GDB	39	43	2	0	97	36	10	1

FIGURE A.4.11. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #7.

1. Phenothiazine (ACN)(8CI)	199 C12H9NS
2. 5H-Indeno[1,2-b]pyridine (8CI9CI)	167 C12H9N
3. Naphtho[1,2-d]thiazole, 2-methyl- (8CI9CI)	199 C12H9NS
4. Naphtho[2,1-d]thiazole, 2-methyl- (8CI9CI)	199 C12H9NS
5. 2-Naphthalenecarboxamide, N,N-dimethyl- (9CI)	199 C13H13NO

Sample file: >L3122 Spectrum #: 1685
Search speed: 2 Tilting option: S No. of ion ranges searched: 55

	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	94*	92842	24266	"B1GDB	102	21	1	0	88	12	64	9
2.	41*	244995	19912	"B1GDB	55	40	2	-1	59	31	16	2
3.	27*	2682453	24269	"B1GDB	39	84	3	0	100	37	10	1
4.	25*	20686628	24276	"B1GDB	26	88	3	0	100	42	8	1
5.	25*	13577850	24253	"B1GDB	24	95	3	0	242	47	7	1

FIGURE A.4.12. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra and Library Hit Spectra of Peak #8.

AIMM

Association for Information and Image Management

1100 Wayne Avenue, Suite 1100
Silver Spring, Maryland 20910
301/587-8202

Centimeter

Inches

MANUFACTURED TO AIMM STANDARDS
BY APPLIED IMAGE, INC.

2 of 3

FIGURE A.4.13. Chloroprene Inhalation Developmental Toxicity Study: Mass Spectra of Broad Peak Between Peak #5 and Peak #6.

PRET	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	1,5-Cyclooctadiene, 1,6-dichloro- (8CI9CI)								176	C8H10Cl2	1	1
2.	Benzenamine, 3-chloro-2-methyl- (9CI)								141	C7H8ClN	1	1
3.	Benzenamine, 2-chloro-6-methyl- (9CI)								141	C7H8ClN	1	1
4.	3,5-Heptadiyn-2-one (8CI9CI)								106	C7H6O	1	1
5.	1-Hexanamine (9CI)								101	C6H15N	1	1
5.	13*	111262	84	"BIGDB	49	26	1	2	67	64	3	3

Sample file: ARTIFI Spectrum #: -1
Search speed: 2 Tilting option: S No. of ion ranges searched: 54

#ru,PS	Prob.	CAS #	CON #	ROOT	K	DK	#FLG	TILT	%	CON	C_I	R_I
1.	60	29480420	7092	"BIGDB	62	53	1	1	100	14	30	1
2.	25*	87605	15957	"BIGDB	43	73	2	2	55	50	7	1
3.	24*	87638	15958	"BIGDB	38	75	2	1	64	41	8	1
4.	24*	13879715	2199	"BIGDB	48	37	1	1	28	55	7	2
5.	13*	111262	84	"BIGDB	49	26	1	2	67	64	3	3

FIGURE A.4.14. Chloroprene Inhalation Developmental Toxicity Study: Library Hit Spectra and Results For Peak at 20.73 min. (See Figure A.4.13).

FIGURE A.4.15. Chloroprene Inhalation Developmental Toxicity Study: GC/MS Total Ion Chromatogram of Sample 53436-72-2.

FIGURE A.4.16. Chloroprene Inhalation Developmental Toxicity Study:
Extracted Ion Chromatogram of Mass 88.

FIGURE A.4.17. Chloroprene Inhalation Developmental Toxicity Study:
Extracted Ion Chromatogram of Mass 176.

APPENDIX B

EXPOSURE DATA

Summation Equations
Concentration Data
Temperature Data
Relative Humidity Data
Exhaust Airflow Data
Exposure Operation Discussion Sheets
Chamber Uniformity Data

Summation Equations

SUMMATION EQUATIONS

Mean: $\bar{X} = \frac{1}{n} \sum_{i=1}^n X_i$

Standard Deviation:

$$S = \sqrt{\frac{\sum_{i=1}^n X_i^2 - \left(\sum_{i=1}^n X_i\right)^2 / n}{n-1}}$$

where:

X_i = individual reading of concentration, temperature or relative humidity

n = number of individual readings

The weekly and study means and standard deviations for concentration were derived from the daily means and standard deviations using the following equation.

Mean: $\bar{X} = \frac{\sum_{j=1}^K (n_j)(\bar{X}_j)}{\sum_{j=1}^K n_j}$

Standard Deviation:

$$S = \sqrt{\frac{\sum_{j=1}^K (n_j - 1) (S_j^2)}{\sum_{j=1}^K n_j - 1}}$$

where:

n_j = number of daily readings

$\bar{X}_j$ = daily mean

S_j = daily standard deviation

K = number of days included in summations

Concentration Data

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 26 Mar 1990 through 19 Apr 1990

Summary Data for: Room		/Concentration		Range=0.00E+0 to 5.00E-1			
Date	Mean	Std Dev	Maximum	Minimum	N	N in	% N in
26 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
27 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
28 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	26	26	100.0%
29 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
30 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
31 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
1 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
2 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
3 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	6	6	100.0%
4 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
5 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
6 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
7 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
8 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
9 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
10 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
11 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
12 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
13 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
14 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	14	14	100.0%
15 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
16 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
17 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
18 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
19 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
Summary	0.00E+00	0.000E+00	0.00E+00	0.00E+00	272	272	100.0%

Chloroprene Develop. Toxicity (Rabbits)
Room
Mean & Standard Deviation
From 26 Mar 1990 through 19 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 0 ppm (A)

/Concentration

Range=0.00E+0 to 5.00E-1

Date	Mean	Std Dev	Maximum	Minimum	N	N in	% N in
27 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
28 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	27	27	100.0%
29 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
30 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
31 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
1 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
2 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	6	6	100.0%
3 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	6	6	100.0%
4 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
5 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
6 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
7 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
8 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
9 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
10 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
11 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
12 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
13 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
14 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	14	14	100.0%
15 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
16 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
17 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
18 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
19 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
Summary	0.00E+00	0.000E+00	0.00E+00	0.00E+00	261	261	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 26 Mar 1990 through 18 Apr 1990

Summary Data for: 0 ppm (B)

/Concentration

Range=0.00E+0 to 5.00E-1

Date	Mean	Std Dev	Maximum	Minimum	N	N in	% N in
26 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
27 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	14	14	100.0%
28 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	24	24	100.0%
29 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	7	7	100.0%
30 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	7	7	100.0%
31 Mar 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
1 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
2 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	7	7	100.0%
3 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	5	5	100.0%
4 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
5 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
6 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	9	9	100.0%
7 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
8 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
9 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	8	8	100.0%
10 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	10	10	100.0%
11 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
12 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
13 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
14 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
15 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	11	11	100.0%
16 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
17 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	12	12	100.0%
18 Apr 1990	0.00E+00	0.000E+00	0.00E+00	0.00E+00	13	13	100.0%
Summary	0.00E+00	0.000E+00	0.00E+00	0.00E+00	261	261	100.0%

Chloroprene Develop. Toxicity (Rabbits)
0 ppm (B)

Mean & Standard Deviation

From 26 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 26 Mar 1990 through 18 Apr 1990

Summary Data for: 10 ppm (A)

/Concentration

Range=9.00E+0 to 1.10E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
26 Mar 1990	9.88E+00	98.8%	1.570E-01	1.6%	1.00E+01	9.55E+00	10	10	100.0%
27 Mar 1990	9.67E+00	96.7%	2.665E-01	2.8%	9.92E+00	9.21E+00	10	10	100.0%
28 Mar 1990	9.57E+00	95.7%	2.225E-01	2.3%	9.97E+00	9.32E+00	11	11	100.0%
29 Mar 1990	9.92E+00	99.2%	6.518E-02	.7%	9.98E+00	9.83E+00	4	4	100.0%
30 Mar 1990	9.59E+00	95.9%	2.467E-02	.3%	9.63E+00	9.56E+00	8	8	100.0%
31 Mar 1990	9.65E+00	96.5%	4.955E-02	.5%	9.72E+00	9.57E+00	8	8	100.0%
1 Apr 1990	9.85E+00	98.5%	4.125E-02	.4%	9.90E+00	9.77E+00	8	8	100.0%
2 Apr 1990	9.78E+00	97.8%	2.003E-01	2.0%	9.87E+00	9.37E+00	6	6	100.0%
3 Apr 1990	9.98E+00	99.8%	1.214E-02	.1%	9.99E+00	9.96E+00	6	6	100.0%
4 Apr 1990	9.93E+00	99.3%	1.686E-02	.2%	9.96E+00	9.91E+00	6	6	100.0%
5 Apr 1990	9.60E+00	96.0%	6.722E-02	.7%	9.70E+00	9.49E+00	7	7	100.0%
6 Apr 1990	9.70E+00	97.0%	9.955E-02	1.0%	9.86E+00	9.54E+00	7	7	100.0%
7 Apr 1990	9.75E+00	97.5%	1.289E-01	1.3%	9.86E+00	9.45E+00	9	9	100.0%
8 Apr 1990	9.88E+00	98.8%	4.435E-02	.4%	9.94E+00	9.83E+00	9	9	100.0%
9 Apr 1990	9.77E+00	97.7%	4.054E-02	.4%	9.81E+00	9.69E+00	7	7	100.0%
10 Apr 1990	1.00E+01	100.4%	1.355E-01	1.3%	1.02E+01	9.76E+00	8	8	100.0%
11 Apr 1990	9.99E+00	99.9%	9.007E-02	.9%	1.01E+01	9.76E+00	9	9	100.0%
12 Apr 1990	9.93E+00	99.3%	2.704E-02	.3%	9.97E+00	9.89E+00	9	9	100.0%
13 Apr 1990	9.85E+00	98.5%	3.574E-02	.4%	9.90E+00	9.77E+00	9	9	100.0%
14 Apr 1990	9.88E+00	98.8%	1.528E-01	1.5%	1.01E+01	9.47E+00	11	11	100.0%
15 Apr 1990	9.89E+00	98.9%	1.285E-01	1.3%	1.01E+01	9.64E+00	8	8	100.0%
16 Apr 1990	9.77E+00	97.7%	1.196E-01	1.2%	9.87E+00	9.51E+00	10	10	100.0%
17 Apr 1990	9.89E+00	98.9%	1.926E-01	1.9%	1.01E+01	9.57E+00	11	11	100.0%
18 Apr 1990	9.98E+00	99.8%	5.645E-02	.6%	1.00E+01	9.82E+00	11	11	100.0%
Summary	9.82E+00	98.2%	1.813E-01	1.8%	1.02E+01	9.21E+00	202	202	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 10 ppm (B)

/Concentration

Range=9.00E+0 to 1.10E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	1.01E+01	101.3%	1.565E-01	1.5%	1.04E+01	9.92E+00	10	10	100.0%
28 Mar 1990	1.00E+01	100.2%	3.309E-01	3.3%	1.05E+01	9.28E+00	11	11	100.0%
29 Mar 1990	1.02E+01	102.1%	2.575E-01	2.5%	1.05E+01	9.87E+00	4	4	100.0%
30 Mar 1990	9.42E+00	94.2%	6.754E-02	.7%	9.49E+00	9.27E+00	8	8	100.0%
31 Mar 1990	9.58E+00	95.8%	1.123E-01	1.2%	9.75E+00	9.38E+00	8	8	100.0%
1 Apr 1990	9.93E+00	99.3%	1.791E-01	1.8%	1.02E+01	9.66E+00	8	8	100.0%
2 Apr 1990	9.88E+00	98.8%	2.335E-01	2.4%	1.02E+01	9.60E+00	5	5	100.0%
3 Apr 1990	1.01E+01	100.8%	2.221E-01	2.2%	1.03E+01	9.80E+00	7	7	100.0%
4 Apr 1990	9.95E+00	99.5%	1.567E-01	1.6%	1.02E+01	9.76E+00	7	7	100.0%
5 Apr 1990	9.76E+00	97.6%	1.674E-02	.2%	9.79E+00	9.75E+00	6	6	100.0%
6 Apr 1990	9.72E+00	97.2%	1.548E-01	1.6%	9.83E+00	9.39E+00	7	7	100.0%
7 Apr 1990	9.90E+00	99.0%	1.784E-01	1.8%	1.01E+01	9.58E+00	9	9	100.0%
8 Apr 1990	9.95E+00	99.5%	2.936E-02	.3%	1.00E+01	9.91E+00	9	9	100.0%
9 Apr 1990	1.02E+01	101.8%	3.716E-02	.4%	1.02E+01	1.01E+01	7	7	100.0%
10 Apr 1990	9.85E+00	98.5%	5.126E-02	.5%	9.91E+00	9.77E+00	8	8	100.0%
11 Apr 1990	9.94E+00	99.4%	3.611E-02	.4%	1.00E+01	9.89E+00	9	9	100.0%
12 Apr 1990	9.92E+00	99.2%	1.976E-02	.2%	9.95E+00	9.89E+00	9	9	100.0%
13 Apr 1990	9.90E+00	99.0%	2.469E-02	.2%	9.94E+00	9.86E+00	9	9	100.0%
14 Apr 1990	9.99E+00	99.9%	1.361E-01	1.4%	1.02E+01	9.79E+00	11	11	100.0%
15 Apr 1990	9.90E+00	99.1%	6.538E-02	.7%	9.95E+00	9.76E+00	8	8	100.0%
16 Apr 1990	1.00E+01	100.4%	7.638E-02	.8%	1.01E+01	9.83E+00	10	10	100.0%
17 Apr 1990	1.01E+01	101.0%	4.315E-02	.4%	1.02E+01	1.01E+01	11	11	100.0%
18 Apr 1990	1.01E+01	100.6%	4.765E-02	.5%	1.01E+01	9.95E+00	11	11	100.0%
19 Apr 1990	1.02E+01	101.9%	4.296E-02	.4%	1.02E+01	1.01E+01	10	10	100.0%
Summary	9.95E+00	99.5%	2.181E-01	2.2%	1.05E+01	9.27E+00	202	202	100.0%

Chloroprene Develop. Toxicity (Rabbits)
10 ppm (B)
Mean & Standard Deviation
From 27 Mar 1990 through 19 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 26 Mar 1990 through 18 Apr 1990

Summary Data for: 40 ppm (A)

/Concentration

Range=3.60E+1 to 4.40E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
26 Mar 1990	3.92E+01	98.1%	8.761E-01	2.2%	3.98E+01	3.73E+01	8	8	100.0%
27 Mar 1990	3.91E+01	97.9%	3.749E-01	1.0%	3.94E+01	3.83E+01	9	9	100.0%
28 Mar 1990	3.99E+01	99.8%	2.705E-01	.7%	4.03E+01	3.94E+01	10	10	100.0%
29 Mar 1990	4.06E+01	101.6%	1.305E-01	.3%	4.08E+01	4.05E+01	4	4	100.0%
30 Mar 1990	3.82E+01	95.5%	1.718E-01	.4%	3.84E+01	3.79E+01	8	8	100.0%
31 Mar 1990	3.85E+01	96.3%	5.420E-01	1.4%	3.92E+01	3.75E+01	8	8	100.0%
1 Apr 1990	4.01E+01	100.3%	4.018E-01	1.0%	4.05E+01	3.92E+01	8	8	100.0%
2 Apr 1990	4.00E+01	99.9%	1.494E-01	.4%	4.01E+01	3.98E+01	5	5	100.0%
3 Apr 1990	3.99E+01	99.8%	1.782E-01	.4%	4.01E+01	3.97E+01	5	5	100.0%
4 Apr 1990	3.98E+01	99.5%	2.481E-01	.6%	4.01E+01	3.95E+01	6	6	100.0%
5 Apr 1990	3.95E+01	98.8%	6.071E-01	1.5%	4.02E+01	3.85E+01	7	7	100.0%
6 Apr 1990	3.97E+01	99.2%	4.724E-01	1.2%	4.00E+01	3.67E+01	7	7	100.0%
7 Apr 1990	3.97E+01	99.3%	2.579E-01	.6%	4.01E+01	3.54E+01	9	9	100.0%
8 Apr 1990	3.95E+01	98.8%	6.145E-01	1.6%	4.00E+01	3.80E+01	9	9	100.0%
9 Apr 1990	3.99E+01	99.8%	3.035E-01	.8%	4.02E+01	3.92E+01	8	8	100.0%
10 Apr 1990	3.96E+01	99.1%	4.904E-01	1.2%	4.01E+01	3.88E+01	8	8	100.0%
11 Apr 1990	3.99E+01	99.6%	3.861E-01	1.0%	4.01E+01	3.89E+01	9	9	100.0%
12 Apr 1990	3.95E+01	98.8%	5.608E-01	1.4%	3.99E+01	3.81E+01	9	9	100.0%
13 Apr 1990	3.91E+01	97.8%	5.883E-01	1.5%	3.94E+01	3.76E+01	9	9	100.0%
14 Apr 1990	3.93E+01	98.3%	5.797E-01	1.5%	4.01E+01	3.85E+01	10	10	100.0%
15 Apr 1990	3.97E+01	99.3%	6.144E-01	1.5%	4.02E+01	3.87E+01	7	7	100.0%
16 Apr 1990	4.04E+01	100.9%	8.352E-01	2.1%	4.07E+01	3.81E+01	9	9	100.0%
17 Apr 1990	3.91E+01	97.8%	6.014E-01	1.5%	4.04E+01	3.83E+01	10	10	100.0%
18 Apr 1990	3.93E+01	98.3%	6.687E-02	.2%	3.94E+01	3.93E+01	10	10	100.0%
Summary	3.95E+01	98.8%	6.860E-01	1.7%	4.08E+01	3.73E+01	192	192	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 40 ppm (B)

/Concentration

Range=3.60E+1 to 4.40E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	3.92E+01	97.9%	4.901E-01	1.3%	3.94E+01	3.78E+01	10	10	100.0%
28 Mar 1990	4.04E+01	101.1%	1.560E+00	3.9%	4.24E+01	3.75E+01	11	11	100.0%
29 Mar 1990	4.06E+01	101.6%	3.092E-01	.8%	4.11E+01	4.04E+01	4	4	100.0%
30 Mar 1990	3.83E+01	95.8%	7.906E-02	.2%	3.84E+01	3.82E+01	8	8	100.0%
31 Mar 1990	3.87E+01	96.6%	2.483E-01	.6%	3.92E+01	3.83E+01	8	8	100.0%
1 Apr 1990	3.97E+01	99.3%	1.800E-01	.5%	3.99E+01	3.94E+01	8	8	100.0%
2 Apr 1990	3.98E+01	99.5%	1.837E-01	.5%	4.01E+01	3.96E+01	6	6	100.0%
3 Apr 1990	3.96E+01	98.9%	2.009E-01	.5%	3.98E+01	3.93E+01	5	5	100.0%
4 Apr 1990	3.94E+01	98.5%	4.170E-01	1.1%	3.97E+01	3.88E+01	6	6	100.0%
5 Apr 1990	3.86E+01	96.6%	2.299E-01	.6%	3.89E+01	3.83E+01	7	7	100.0%
6 Apr 1990	3.88E+01	97.0%	5.284E-01	1.4%	3.94E+01	3.78E+01	7	7	100.0%
7 Apr 1990	3.99E+01	99.8%	1.231E+00	3.1%	4.14E+01	3.82E+01	9	9	100.0%
8 Apr 1990	4.05E+01	101.3%	4.004E-01	1.0%	4.10E+01	3.97E+01	9	9	100.0%
9 Apr 1990	4.00E+01	100.0%	8.831E-01	2.2%	4.16E+01	3.94E+01	7	7	100.0%
10 Apr 1990	3.96E+01	98.9%	2.974E-01	.8%	3.99E+01	3.91E+01	8	8	100.0%
11 Apr 1990	3.93E+01	98.2%	2.561E-01	.7%	3.96E+01	3.88E+01	9	9	100.0%
12 Apr 1990	3.90E+01	97.5%	4.879E-01	1.3%	4.02E+01	3.86E+01	9	9	100.0%
13 Apr 1990	4.03E+01	100.9%	2.426E-01	.6%	4.08E+01	3.99E+01	9	9	100.0%
14 Apr 1990	4.02E+01	100.4%	8.268E-01	2.1%	4.16E+01	3.89E+01	11	11	100.0%
15 Apr 1990	4.01E+01	100.2%	5.503E-01	1.4%	4.11E+01	3.95E+01	8	8	100.0%
16 Apr 1990	3.96E+01	99.0%	3.798E-01	1.0%	4.00E+01	3.88E+01	10	10	100.0%
17 Apr 1990	3.98E+01	99.5%	3.604E-01	.9%	4.01E+01	3.88E+01	11	11	100.0%
18 Apr 1990	3.95E+01	98.8%	4.956E-01	1.3%	3.99E+01	3.82E+01	11	11	100.0%
19 Apr 1990	4.00E+01	100.0%	6.393E-01	1.6%	4.03E+01	3.82E+01	10	10	100.0%
Summary	3.96E+01	99.1%	8.406E-01	2.1%	4.24E+01	3.75E+01	201	201	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 26 Mar 1990 through 18 Apr 1990

Summary Data for: 175 ppm (A)

/Concentration

Range=1.58E+2 to 1.93E+2

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
26 Mar 1990	1.75E+02	100.3%	3.625E-01	.2%	1.76E+02	1.75E+02	8	8	100.0%
27 Mar 1990	1.75E+02	99.9%	3.598E-01	.2%	1.76E+02	1.74E+02	9	9	100.0%
28 Mar 1990	1.76E+02	100.5%	1.141E+00	.6%	1.77E+02	1.75E+02	10	10	100.0%
29 Mar 1990	1.84E+02	105.2%	3.139E+00	1.7%	1.87E+02	1.81E+02	3	3	100.0%
30 Mar 1990	1.70E+02	97.2%	5.357E-01	.3%	1.71E+02	1.69E+02	8	8	100.0%
31 Mar 1990	1.72E+02	98.1%	4.000E-01	.2%	1.72E+02	1.71E+02	7	7	100.0%
1 Apr 1990	1.74E+02	99.3%	6.266E-01	.4%	1.74E+02	1.73E+02	8	8	100.0%
2 Apr 1990	1.74E+02	99.2%	2.168E-01	.1%	1.74E+02	1.73E+02	5	5	100.0%
3 Apr 1990	1.74E+02	99.4%	5.164E-02	0.0%	1.74E+02	1.74E+02	6	6	100.0%
4 Apr 1990	1.74E+02	99.6%	3.266E-01	.2%	1.75E+02	1.74E+02	6	6	100.0%
5 Apr 1990	1.78E+02	101.7%	1.441E+00	.8%	1.80E+02	1.76E+02	6	6	100.0%
6 Apr 1990	1.76E+02	100.6%	6.451E-01	.4%	1.77E+02	1.75E+02	7	7	100.0%
7 Apr 1990	1.77E+02	101.4%	3.227E-01	.2%	1.78E+02	1.77E+02	8	8	100.0%
8 Apr 1990	1.76E+02	100.6%	5.069E-01	.3%	1.77E+02	1.76E+02	8	8	100.0%
9 Apr 1990	1.76E+02	100.5%	1.066E+00	.6%	1.78E+02	1.75E+02	7	7	100.0%
10 Apr 1990	1.77E+02	101.1%	1.127E+00	.6%	1.79E+02	1.76E+02	7	7	100.0%
11 Apr 1990	1.77E+02	101.1%	5.155E-01	.3%	1.78E+02	1.76E+02	8	8	100.0%
12 Apr 1990	1.77E+02	100.9%	8.288E-01	.5%	1.78E+02	1.76E+02	8	8	100.0%
13 Apr 1990	1.77E+02	100.9%	5.451E-01	.3%	1.78E+02	1.76E+02	8	8	100.0%
14 Apr 1990	1.77E+02	101.2%	4.695E+00	2.7%	1.85E+02	1.72E+02	10	10	100.0%
15 Apr 1990	1.76E+02	100.4%	5.159E-01	.3%	1.76E+02	1.75E+02	7	7	100.0%
16 Apr 1990	1.75E+02	100.2%	6.654E-01	.4%	1.77E+02	1.75E+02	10	10	100.0%
17 Apr 1990	1.74E+02	99.4%	8.781E-01	.5%	1.75E+02	1.72E+02	10	10	100.0%
18 Apr 1990	1.75E+02	100.2%	1.829E-01	.1%	1.76E+02	1.75E+02	10	10	100.0%
Summary	1.75E+02	100.3%	2.452E+00	1.4%	1.87E+02	1.69E+02	184	184	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 175 ppm (B)

/Concentration

Range=1.58E+2 to 1.93E+2

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	1.74E+02	99.6%	2.991E-01	.2%	1.75E+02	1.74E+02	9	9	100.0%
28 Mar 1990	1.75E+02	100.1%	8.720E-01	.5%	1.78E+02	1.75E+02	10	10	100.0%
29 Mar 1990	1.76E+02	100.4%	2.061E+00	1.2%	1.77E+02	1.73E+02	4	4	100.0%
30 Mar 1990	1.73E+02	98.8%	1.332E+00	.8%	1.75E+02	1.72E+02	8	8	100.0%
31 Mar 1990	1.74E+02	99.4%	2.416E-01	.1%	1.74E+02	1.74E+02	8	8	100.0%
1 Apr 1990	1.75E+02	99.9%	3.055E-01	.2%	1.75E+02	1.75E+02	7	7	100.0%
2 Apr 1990	1.77E+02	100.9%	8.367E-02	0.0%	1.77E+02	1.76E+02	5	5	100.0%
3 Apr 1990	1.75E+02	100.2%	3.114E-01	.2%	1.76E+02	1.75E+02	5	5	100.0%
4 Apr 1990	1.75E+02	99.9%	3.189E-01	.2%	1.76E+02	1.75E+02	6	6	100.0%
5 Apr 1990	1.74E+02	99.7%	1.271E+00	.7%	1.76E+02	1.73E+02	6	6	100.0%
6 Apr 1990	1.72E+02	98.0%	7.139E-01	.4%	1.73E+02	1.71E+02	6	6	100.0%
7 Apr 1990	1.75E+02	100.0%	1.263E+00	.7%	1.76E+02	1.72E+02	8	8	100.0%
8 Apr 1990	1.75E+02	100.0%	5.339E-01	.3%	1.76E+02	1.74E+02	8	8	100.0%
9 Apr 1990	1.75E+02	99.8%	1.545E+00	.9%	1.78E+02	1.74E+02	7	7	100.0%
10 Apr 1990	1.75E+02	99.8%	2.029E+00	1.2%	1.79E+02	1.74E+02	7	7	100.0%
11 Apr 1990	1.76E+02	100.6%	2.020E+00	1.1%	1.81E+02	1.75E+02	8	8	100.0%
12 Apr 1990	1.74E+02	99.3%	2.073E+00	1.2%	1.79E+02	1.72E+02	8	8	100.0%
13 Apr 1990	1.75E+02	100.0%	5.097E-01	.3%	1.76E+02	1.74E+02	8	8	100.0%
14 Apr 1990	1.75E+02	100.2%	2.358E+00	1.3%	1.80E+02	1.73E+02	10	10	100.0%
15 Apr 1990	1.77E+02	101.1%	7.319E-01	.4%	1.78E+02	1.76E+02	7	7	100.0%
16 Apr 1990	1.74E+02	99.2%	1.270E+00	.7%	1.75E+02	1.70E+02	10	10	100.0%
17 Apr 1990	1.74E+02	99.2%	1.541E+00	.9%	1.76E+02	1.70E+02	10	10	100.0%
18 Apr 1990	1.75E+02	100.2%	8.486E-01	.5%	1.76E+02	1.74E+02	10	10	100.0%
19 Apr 1990	1.76E+02	100.7%	4.206E-01	.2%	1.77E+02	1.76E+02	9	9	100.0%
Summary	1.75E+02	99.9%	1.622E+00	.9%	1.81E+02	1.70E+02	184	184	100.0%

Chloroprene Develop. Toxicity (Rabbits)

175 ppm (B)

Mean & Standard Deviation

From 27 Mar 1990 through 19 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 26 Mar 1990 through 26 Mar 1990

Summary Data for: Standard Gas

/Concentration

Range=4.14E+0 to 5.06E+0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
26 Mar 1990	4.26E+00	92.7%	5.173E-02	1.2%	4.34E+00	4.21E+00	9	9	100.0%
Summary	4.26E+00	92.7%	5.173E-02	1.2%	4.34E+00	4.21E+00	9	9	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 31 Mar 1990

Summary Data for: Standard Gas

/Concentration

Range=2.70E+1 to 3.30E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	3.19E+01	106.2%	6.651E-02	.2%	3.19E+01	3.17E+01	12	12	100.0%
28 Mar 1990	3.20E+01	106.6%	9.477E-02	.3%	3.21E+01	3.19E+01	11	11	100.0%
29 Mar 1990	3.30E+01	109.8%	1.415E+00	4.3%	3.55E+01	3.22E+01	5	4	80.0%
30 Mar 1990	3.54E+01	117.9%	7.770E-01	2.2%	3.60E+01	3.35E+01	8	0	0.0%
31 Mar 1990	3.57E+01	119.1%	1.502E-01	.4%	3.59E+01	3.54E+01	8	0	0.0%
Summary	3.34E+01	111.2%	1.791E+00	5.4%	3.60E+01	3.17E+01	44	27	61.4%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 1 Apr 1990 through 7 Apr 1990

Summary Data for: Standard Gas

/Concentration

Range=2.97E+1 to 3.63E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
1 Apr 1990	3.59E+01	108.7%	9.996E-02	.3%	3.60E+01	3.57E+01	8	8	100.0%
2 Apr 1990	3.59E+01	108.9%	7.189E-01	2.0%	3.64E+01	3.45E+01	6	4	66.7%
3 Apr 1990	3.64E+01	110.2%	1.103E+00	3.0%	3.68E+01	3.31E+01	11	1	9.1%
4 Apr 1990	3.65E+01	110.5%	8.361E-02	.2%	3.66E+01	3.63E+01	7	0	0.0%
5 Apr 1990	3.63E+01	110.1%	1.217E-01	.3%	3.65E+01	3.61E+01	7	3	42.9%
6 Apr 1990	3.67E+01	111.1%	4.488E-01	1.2%	3.78E+01	3.62E+01	9	1	11.1%
7 Apr 1990	3.62E+01	109.7%	1.034E-01	.3%	3.64E+01	3.60E+01	9	8	88.9%
Summary	3.63E+01	109.9%	8.025E-01	1.7%	3.78E+01	3.31E+01	57	25	43.9%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 8 Apr 1990 through 19 Apr 1990

Summary Data for: Standard Gas

/Concentration

Range=3.27E+1 to 3.99E+1

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
8 Apr 1990	3.65E+01	100.5%	1.556E-01	.4%	3.68E+01	3.62E+01	8	8	100.0
9 Apr 1990	3.74E+01	102.9%	8.887E-02	.2%	3.75E+01	3.72E+01	8	8	100.0
10 Apr 1990	3.76E+01	103.6%	1.586E-01	.4%	3.82E+01	3.74E+01	15	15	100.0
11 Apr 1990	3.72E+01	102.4%	1.385E-01	.4%	3.73E+01	3.69E+01	9	9	100.0
12 Apr 1990	3.71E+01	102.1%	1.413E-01	.4%	3.72E+01	3.68E+01	9	9	100.0
13 Apr 1990	3.69E+01	101.8%	1.328E-01	.4%	3.71E+01	3.68E+01	9	9	100.0
14 Apr 1990	3.69E+01	101.8%	8.256E-02	.2%	3.70E+01	3.68E+01	11	11	100.0
15 Apr 1990	3.65E+01	100.7%	1.245E-01	.3%	3.67E+01	3.63E+01	10	10	100.0
16 Apr 1990	3.64E+01	100.3%	5.254E-02	.1%	3.65E+01	3.63E+01	10	10	100.0
17 Apr 1990	3.65E+01	100.5%	1.251E-01	.3%	3.67E+01	3.62E+01	10	10	100.0
18 Apr 1990	3.68E+01	101.4%	1.663E-01	.5%	3.71E+01	3.65E+01	11	11	100.0
19 Apr 1990	3.69E+01	101.6%	2.238E-01	.6%	3.73E+01	3.65E+01	10	10	100.0
Summary	3.69E+01	101.7%	4.025E-01	1.1%	3.82E+01	3.62E+01	120	120	100.0

Chloroprene Develop. Toxicity (Rabbits)
 Standard Gas
 Mean & Standard Deviation
 From 26 Mar 1990 through 19 Apr 1990

Temperature Data

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: Room

/Temperature

Range= 61.0 to 69.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	62.2	95.7%	2.24	3.6%	65.3	59.7	7	4	57.1%
24 Mar 1990	64.1	98.7%	1.14	1.8%	65.9	62.1	8	8	100.0%
25 Mar 1990	64.2	98.8%	.37	.6%	64.6	63.6	8	8	100.0%
26 Mar 1990	64.2	98.7%	.48	.7%	64.6	63.1	7	7	100.0%
27 Mar 1990	64.6	99.4%	.53	.8%	65.1	63.8	8	8	100.0%
28 Mar 1990	61.9	95.2%	2.10	3.4%	64.6	58.5	8	5	62.5%
29 Mar 1990	62.2	95.7%	1.15	1.8%	64.0	60.7	7	5	71.4%
30 Mar 1990	63.2	97.3%	.60	1.0%	64.2	62.5	8	8	100.0%
31 Mar 1990	62.8	96.6%	.31	.5%	63.2	62.4	8	8	100.0%
1 Apr 1990	64.0	98.5%	1.08	1.7%	65.8	62.3	8	8	100.0%
2 Apr 1990	63.9	98.3%	.80	1.3%	65.3	63.0	7	7	100.0%
3 Apr 1990	63.1	97.1%	.25	.4%	63.4	62.7	8	8	100.0%
4 Apr 1990	63.0	96.9%	.19	.3%	63.3	62.8	7	7	100.0%
5 Apr 1990	63.7	98.0%	.75	1.2%	65.0	62.7	7	7	100.0%
6 Apr 1990	63.3	97.4%	.79	1.2%	64.7	62.3	7	7	100.0%
7 Apr 1990	62.7	96.4%	.45	.7%	63.2	62.1	8	8	100.0%
8 Apr 1990	62.3	95.8%	.41	.7%	62.6	61.7	7	7	100.0%
9 Apr 1990	63.9	98.3%	.77	1.2%	64.6	62.4	8	8	100.0%
10 Apr 1990	64.7	99.5%	.50	.8%	65.5	64.1	6	6	100.0%
11 Apr 1990	65.2	100.3%	.56	.9%	65.7	64.3	8	8	100.0%
12 Apr 1990	66.0	101.6%	.84	1.3%	66.9	64.7	7	7	100.0%
13 Apr 1990	65.6	101.0%	.73	1.1%	66.5	64.3	8	8	100.0%
14 Apr 1990	66.0	101.5%	.42	.6%	66.4	65.3	7	7	100.0%
15 Apr 1990	67.3	103.6%	.77	1.1%	67.9	65.5	8	8	100.0%
16 Apr 1990	67.3	103.6%	.46	.7%	67.9	66.7	8	8	100.0%
17 Apr 1990	66.4	102.1%	.66	1.0%	67.1	65.3	8	8	100.0%
18 Apr 1990	65.9	101.5%	.43	.7%	66.5	65.2	7	7	100.0%
Summary	64.2	98.8%	1.75	2.7%	67.9	58.5	203	195	96.1%

Chloroprene Develop. Toxicity (Rabbits)
Room

Mean & Standard Deviation
From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 0 ppm (A)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	66.8	96.8%	.49	.7%	67.4	66.1	5	5	100.0%
28 Mar 1990	67.2	97.3%	1.32	2.0%	69.3	65.4	7	7	100.0%
29 Mar 1990	66.6	96.5%	.67	1.0%	67.3	65.2	7	7	100.0%
30 Mar 1990	67.4	97.6%	.89	1.3%	68.9	66.3	8	8	100.0%
31 Mar 1990	67.6	97.9%	.67	1.0%	68.8	66.6	7	7	100.0%
1 Apr 1990	68.1	98.6%	.80	1.2%	69.1	67.3	6	6	100.0%
2 Apr 1990	67.1	97.2%	.37	.6%	67.6	66.6	6	6	100.0%
3 Apr 1990	66.9	96.9%	.56	.8%	67.6	66.0	8	8	100.0%
4 Apr 1990	66.6	96.6%	.63	.9%	67.3	65.3	7	7	100.0%
5 Apr 1990	67.5	97.8%	.24	.4%	67.9	67.2	6	6	100.0%
6 Apr 1990	66.9	97.0%	.56	.8%	67.4	65.8	7	7	100.0%
7 Apr 1990	66.7	96.6%	.18	.3%	66.9	66.4	7	7	100.0%
8 Apr 1990	66.7	96.7%	.29	.4%	67.0	66.4	6	6	100.0%
9 Apr 1990	66.5	96.3%	.49	.7%	67.4	66.0	8	8	100.0%
10 Apr 1990	66.5	96.3%	.53	.8%	67.0	65.7	6	6	100.0%
11 Apr 1990	67.4	97.6%	.55	.8%	68.2	66.6	7	7	100.0%
12 Apr 1990	67.7	98.0%	.36	.5%	68.2	67.2	6	6	100.0%
13 Apr 1990	67.8	98.2%	.39	.6%	68.3	67.3	7	7	100.0%
14 Apr 1990	67.7	98.1%	.30	.4%	68.0	67.2	7	7	100.0%
15 Apr 1990	67.6	98.0%	.19	.3%	68.0	67.4	8	8	100.0%
16 Apr 1990	68.3	98.9%	.36	.5%	68.6	67.7	7	7	100.0%
17 Apr 1990	68.0	98.6%	.30	.4%	68.3	67.4	8	8	100.0%
18 Apr 1990	68.0	98.6%	.37	.5%	68.6	67.4	7	7	100.0%
19 Apr 1990	66.9	97.0%	.49	.7%	67.3	66.2	7	7	100.0%
Summary	67.3	97.5%	.76	1.1%	69.3	65.2	165	165	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 0 ppm (B)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	65.1	94.4%	1.93	3.0%	68.6	62.9	7	6	85.7%
24 Mar 1990	66.9	96.9%	1.19	1.8%	68.4	64.4	8	8	100.0%
25 Mar 1990	67.4	97.7%	.97	1.4%	68.9	66.3	8	8	100.0%
26 Mar 1990	67.8	98.3%	.65	1.0%	68.3	66.5	7	7	100.0%
27 Mar 1990	68.8	99.7%	.81	1.2%	69.7	67.2	7	7	100.0%
28 Mar 1990	67.3	97.5%	1.46	2.2%	69.1	64.9	7	6	85.7%
29 Mar 1990	66.2	95.9%	.66	1.0%	67.2	65.2	7	7	100.0%
30 Mar 1990	66.7	96.7%	.98	1.5%	68.6	65.5	8	8	100.0%
31 Mar 1990	66.8	96.8%	.68	1.0%	67.9	65.7	8	8	100.0%
1 Apr 1990	67.5	97.9%	.91	1.3%	68.9	66.8	6	6	100.0%
2 Apr 1990	67.0	97.1%	.38	.6%	67.4	66.4	6	6	100.0%
3 Apr 1990	66.6	96.5%	.41	.6%	67.0	65.8	8	8	100.0%
4 Apr 1990	66.4	96.3%	.69	1.0%	67.3	65.4	7	7	100.0%
5 Apr 1990	67.2	97.4%	.28	.4%	67.4	66.7	6	6	100.0%
6 Apr 1990	66.7	96.7%	.58	.9%	67.8	65.8	7	7	100.0%
7 Apr 1990	66.7	96.7%	.44	.7%	67.5	66.3	7	7	100.0%
8 Apr 1990	66.4	96.2%	.39	.6%	67.0	65.8	6	6	100.0%
9 Apr 1990	66.6	96.5%	.40	.6%	67.0	65.8	8	8	100.0%
10 Apr 1990	66.6	96.5%	.40	.6%	67.0	66.0	6	6	100.0%
11 Apr 1990	67.6	98.0%	.42	.6%	68.2	67.1	7	7	100.0%
12 Apr 1990	67.9	98.4%	.24	.4%	68.3	67.7	6	6	100.0%
13 Apr 1990	67.8	98.3%	.24	.4%	68.2	67.4	8	8	100.0%
14 Apr 1990	67.9	98.4%	.29	.4%	68.1	67.3	7	7	100.0%
15 Apr 1990	67.8	98.3%	.29	.4%	68.2	67.4	8	8	100.0%
16 Apr 1990	68.1	98.7%	.16	.2%	68.4	67.9	7	7	100.0%
17 Apr 1990	68.1	98.6%	.27	.4%	68.5	67.7	8	8	100.0%
18 Apr 1990	68.1	95.7%	.53	.8%	67.2	65.6	7	7	100.0%
Summary	67.1	97.3%	1.04	1.6%	69.7	62.9	192	190	99.0%

Chloroprene Develop. Toxicity (Rabbits)

0 ppm (B)

Mean & Standard Deviation

From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 10 ppm (A)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	63.4	91.9%	2.33	3.7%	67.7	60.7	7	4	57.1%
24 Mar 1990	65.5	94.9%	1.44	2.2%	67.2	62.7	8	7	87.5%
25 Mar 1990	65.8	95.4%	.77	1.2%	66.8	64.5	8	8	100.0%
26 Mar 1990	67.0	97.1%	1.04	1.5%	68.0	65.2	7	7	100.0%
27 Mar 1990	68.1	98.7%	.83	1.2%	68.9	66.5	7	7	100.0%
28 Mar 1990	66.2	96.0%	1.93	2.9%	68.6	63.3	7	5	71.4%
29 Mar 1990	65.0	94.2%	1.05	1.6%	68.3	63.1	6	3	50.0%
30 Mar 1990	66.1	95.7%	1.33	2.0%	68.2	63.8	8	7	87.5%
31 Mar 1990	66.9	97.0%	.98	1.5%	68.6	65.7	8	8	100.0%
1 Apr 1990	67.1	97.3%	1.21	1.8%	68.8	66.1	6	6	100.0%
2 Apr 1990	66.6	96.5%	.84	1.0%	67.3	65.7	6	6	100.0%
3 Apr 1990	66.1	95.8%	.90	1.4%	67.2	64.8	8	7	87.5%
4 Apr 1990	65.7	95.2%	.60	.9%	66.6	64.8	7	6	85.7%
5 Apr 1990	66.5	96.4%	.54	.8%	67.5	66.0	6	6	100.0%
6 Apr 1990	66.2	96.0%	1.07	1.6%	67.4	64.7	7	5	71.4%
7 Apr 1990	66.5	96.4%	.57	.9%	67.0	65.5	7	7	100.0%
8 Apr 1990	65.3	94.7%	.58	.9%	66.2	64.8	6	3	50.0%
9 Apr 1990	65.1	94.3%	.55	.8%	66.1	64.2	8	5	62.5%
10 Apr 1990	67.0	97.1%	1.21	1.8%	68.2	64.9	6	5	83.3%
11 Apr 1990	68.3	99.0%	.63	.9%	69.0	67.2	7	7	100.0%
12 Apr 1990	68.9	99.9%	.52	.7%	69.6	68.1	6	6	100.0%
13 Apr 1990	68.9	99.8%	.60	.9%	69.6	68.0	8	8	100.0%
14 Apr 1990	69.0	100.0%	.48	.7%	69.6	68.3	7	7	100.0%
15 Apr 1990	69.2	100.3%	.45	.6%	70.0	68.7	8	8	100.0%
16 Apr 1990	69.0	100.0%	.39	.6%	69.5	68.4	7	7	100.0%
17 Apr 1990	68.9	99.9%	1.00	1.5%	69.9	66.9	7	7	100.0%
18 Apr 1990	67.2	97.4%	.48	.7%	68.1	66.6	7	7	100.0%
Summary	66.9	96.9%	1.76	2.6%	70.0	60.7	190	169	88.9%

Chloroprene Develop. Toxicity (Rabbits)

10 ppm (A)

Mean & Standard Deviation

From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 10 ppm (B)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	67.0	97.1%	.30	.5%	67.3	66.5	6	6	100.0%
28 Mar 1990	67.3	97.5%	1.46	2.2%	69.3	65.2	7	7	100.0%
29 Mar 1990	66.3	96.2%	.49	.7%	66.9	65.4	6	6	100.0%
30 Mar 1990	67.4	97.7%	.96	1.4%	68.9	65.9	8	8	100.0%
31 Mar 1990	67.9	98.4%	.89	1.3%	69.3	66.8	8	8	100.0%
1 Apr 1990	68.1	98.8%	.77	1.1%	69.2	67.4	6	6	100.0%
2 Apr 1990	67.4	97.7%	.43	.6%	68.1	66.8	6	6	100.0%
3 Apr 1990	66.9	97.0%	.24	.4%	67.3	66.6	8	8	100.0%
4 Apr 1990	67.0	97.0%	.36	.5%	67.3	66.5	6	6	100.0%
5 Apr 1990	67.5	97.8%	.28	.4%	67.8	67.0	6	6	100.0%
6 Apr 1990	67.1	97.3%	.57	.9%	67.8	66.2	7	7	100.0%
7 Apr 1990	67.0	97.1%	.44	.7%	67.5	66.4	7	7	100.0%
8 Apr 1990	66.8	96.8%	.26	.4%	67.1	66.5	6	6	100.0%
9 Apr 1990	66.8	96.8%	.39	.6%	67.4	66.3	8	8	100.0%
10 Apr 1990	67.2	97.4%	.33	.5%	67.6	66.7	5	5	100.0%
11 Apr 1990	67.8	98.3%	.48	.7%	68.3	67.1	7	7	100.0%
12 Apr 1990	68.2	98.9%	.34	.5%	68.8	67.8	6	6	100.0%
13 Apr 1990	68.4	99.2%	.61	.9%	69.4	67.3	8	8	100.0%
14 Apr 1990	68.1	98.7%	.24	.4%	68.5	67.7	7	7	100.0%
15 Apr 1990	68.2	98.9%	.37	.5%	69.0	67.7	8	8	100.0%
16 Apr 1990	68.1	98.7%	.26	.4%	68.4	67.7	7	7	100.0%
17 Apr 1990	68.5	99.3%	.52	.8%	69.3	67.9	7	7	100.0%
18 Apr 1990	68.7	99.6%	.41	.6%	69.0	67.8	7	7	100.0%
19 Apr 1990	67.0	97.1%	.43	.6%	67.5	66.3	7	7	100.0%
Summary	67.6	97.9%	.84	1.2%	69.4	65.2	164	164	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 40 ppm (A)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	63.5	92.0%	1.70	2.7%	66.5	61.3	7	4	57.1%
24 Mar 1990	64.8	93.9%	1.39	2.1%	66.5	62.5	8	7	87.5%
25 Mar 1990	65.2	94.4%	1.21	1.9%	66.5	63.8	8	8	100.0%
26 Mar 1990	65.8	95.4%	.98	1.5%	66.8	63.9	7	6	85.7%
27 Mar 1990	67.4	97.6%	1.02	1.5%	68.2	65.2	7	7	100.0%
28 Mar 1990	65.5	95.0%	1.22	1.9%	66.9	63.5	7	5	71.4%
29 Mar 1990	64.5	93.5%	.63	1.0%	65.4	63.7	6	1	16.7%
30 Mar 1990	65.5	95.0%	1.10	1.7%	67.0	63.9	8	6	75.0%
31 Mar 1990	66.0	95.7%	.93	1.4%	67.6	64.8	8	7	87.5%
1 Apr 1990	66.9	96.9%	.94	1.4%	68.1	65.6	6	6	100.0%
2 Apr 1990	65.8	95.3%	.41	.6%	66.3	65.2	6	6	100.0%
3 Apr 1990	65.5	94.9%	.54	.8%	66.5	64.8	8	6	75.0%
4 Apr 1990	65.3	94.6%	.47	.7%	65.7	64.4	6	5	83.3%
5 Apr 1990	65.9	95.5%	.30	.5%	66.3	65.6	6	6	100.0%
6 Apr 1990	65.5	94.9%	.65	1.0%	66.4	64.5	7	6	85.7%
7 Apr 1990	65.4	94.8%	.64	1.0%	66.3	64.5	7	5	71.4%
8 Apr 1990	65.2	94.5%	.51	.8%	65.9	64.5	6	4	66.7%
9 Apr 1990	65.1	94.3%	.55	.8%	65.7	64.1	8	5	62.5%
10 Apr 1990	67.7	98.1%	.19	.3%	67.9	67.5	5	5	100.0%
11 Apr 1990	68.4	99.2%	.90	1.3%	69.6	66.9	7	7	100.0%
12 Apr 1990	69.3	100.4%	.50	.7%	70.1	68.7	6	6	100.0%
13 Apr 1990	69.4	100.6%	.54	.8%	70.1	68.4	8	8	100.0%
14 Apr 1990	69.3	100.4%	.31	.4%	69.7	68.8	7	7	100.0%
15 Apr 1990	69.3	100.5%	.27	.4%	69.9	69.1	8	8	100.0%
16 Apr 1990	69.2	100.2%	.16	.2%	69.4	69.0	7	7	100.0%
17 Apr 1990	69.2	100.3%	.38	.5%	69.9	68.9	7	7	100.0%
18 Apr 1990	67.0	97.1%	.52	.8%	67.3	66.0	6	6	100.0%
Summary	66.6	96.5%	1.92	2.9%	70.1	61.3	187	161	86.1%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 40 ppm (B)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	65.9	95.5%	.20	.3%	66.1	65.6	6	6	100.0%
28 Mar 1990	66.6	96.5%	1.01	1.5%	67.9	65.2	7	7	100.0%
29 Mar 1990	65.3	94.6%	.31	.5%	65.6	64.7	6	5	83.3%
30 Mar 1990	66.9	97.0%	1.05	1.6%	68.4	64.9	8	7	87.5%
31 Mar 1990	67.1	97.3%	.45	.7%	68.0	66.6	8	8	100.0%
1 Apr 1990	67.6	97.9%	.74	1.1%	68.6	66.7	6	6	100.0%
2 Apr 1990	67.1	97.3%	.24	.4%	67.5	66.8	6	6	100.0%
3 Apr 1990	66.8	96.8%	.42	.6%	67.4	66.2	8	8	100.0%
4 Apr 1990	66.9	96.9%	.45	.7%	67.8	66.5	6	6	100.0%
5 Apr 1990	67.5	97.9%	.27	.4%	68.0	67.2	6	6	100.0%
6 Apr 1990	66.9	97.0%	.68	1.0%	67.6	65.6	7	7	100.0%
7 Apr 1990	67.0	97.2%	.36	.5%	67.6	66.6	7	7	100.0%
8 Apr 1990	66.5	96.3%	.19	.3%	66.7	66.2	6	6	100.0%
9 Apr 1990	66.6	96.6%	.42	.6%	67.6	66.3	8	8	100.0%
10 Apr 1990	67.8	98.2%	.27	.4%	68.1	67.4	5	5	100.0%
11 Apr 1990	68.2	98.8%	.71	1.0%	68.9	67.2	7	7	100.0%
12 Apr 1990	68.7	99.6%	.31	.5%	69.3	68.4	6	6	100.0%
13 Apr 1990	68.3	98.9%	.22	.3%	68.6	67.9	8	8	100.0%
14 Apr 1990	68.8	99.7%	.36	.5%	69.3	68.4	7	7	100.0%
15 Apr 1990	68.8	99.7%	.17	.3%	69.1	68.5	8	8	100.0%
16 Apr 1990	68.4	99.2%	.30	.4%	68.8	68.1	7	7	100.0%
17 Apr 1990	68.7	99.5%	.16	.2%	68.9	68.5	7	7	100.0%
18 Apr 1990	68.8	99.7%	.33	.5%	69.2	68.3	6	6	100.0%
19 Apr 1990	68.0	98.5%	.32	.5%	68.4	67.5	7	7	100.0%
Summary	67.5	97.8%	1.06	1.6%	69.3	64.7	163	161	98.8%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 175 ppm (A)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev.	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	65.5	95.0%	1.26	1.9%	67.9	64.3	6	6	100.0%
24 Mar 1990	66.8	96.8%	1.02	1.5%	68.1	65.1	8	8	100.0%
25 Mar 1990	67.3	97.5%	.80	1.2%	68.3	66.1	8	8	100.0%
26 Mar 1990	67.2	97.5%	.57	.8%	67.7	66.2	7	7	100.0%
27 Mar 1990	68.9	99.9%	1.03	1.5%	69.9	66.7	7	7	100.0%
28 Mar 1990	68.0	98.5%	1.01	1.5%	69.0	66.1	7	7	100.0%
29 Mar 1990	67.1	97.2%	.47	.7%	67.6	66.2	6	6	100.0%
30 Mar 1990	67.9	98.4%	.95	1.4%	69.4	66.7	8	8	100.0%
31 Mar 1990	68.1	98.7%	.67	1.0%	69.3	67.5	8	8	100.0%
1 Apr 1990	68.6	99.5%	.96	1.4%	69.9	67.6	6	6	100.0%
2 Apr 1990	67.9	98.4%	.48	.7%	68.5	67.2	6	6	100.0%
3 Apr 1990	67.4	97.7%	.20	.3%	67.8	67.2	8	8	100.0%
4 Apr 1990	67.8	98.3%	.30	.4%	68.3	67.4	6	6	100.0%
5 Apr 1990	68.7	99.6%	.48	.7%	69.5	68.2	6	6	100.0%
6 Apr 1990	68.5	99.2%	.86	1.2%	69.6	67.1	7	7	100.0%
7 Apr 1990	68.2	98.9%	.69	1.0%	69.2	67.6	7	7	100.0%
8 Apr 1990	68.0	98.6%	.40	.6%	68.6	67.6	6	6	100.0%
9 Apr 1990	67.8	98.2%	.54	.8%	68.7	67.2	8	8	100.0%
10 Apr 1990	68.1	98.7%	.66	1.0%	68.9	67.3	5	5	100.0%
11 Apr 1990	68.7	99.6%	.72	1.0%	69.8	67.7	7	7	100.0%
12 Apr 1990	68.2	98.8%	.69	1.0%	68.9	66.9	6	6	100.0%
13 Apr 1990	68.5	99.3%	.41	.6%	69.1	68.1	8	8	100.0%
14 Apr 1990	68.8	99.7%	.32	.5%	69.4	68.4	7	7	100.0%
15 Apr 1990	68.8	99.7%	.33	.5%	69.2	68.4	8	8	100.0%
16 Apr 1990	68.7	99.6%	.33	.5%	69.1	68.2	7	7	100.0%
17 Apr 1990	68.9	99.9%	.43	.6%	69.3	68.2	7	7	100.0%
18 Apr 1990	68.4	98.3%	.68	1.0%	68.9	65.1	6	6	100.0%
Summary	68.0	98.5%	1.03	1.5%	69.9	64.3	186	186	100.0%

Chloroprene Develop. Toxicity (Rabbits)

175 ppm (A)

Mean & Standard Deviation

From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 175 ppm (B)

/Temperature

Range= 65.0 to 73.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	67.4	97.6%	.45	.7%	67.8	66.7	6	6	100.0%
28 Mar 1990	68.0	98.5%	1.43	2.1%	69.6	65.9	6	6	100.0%
29 Mar 1990	67.0	97.1%	.68	1.0%	67.6	65.7	6	6	100.0%
30 Mar 1990	67.7	98.2%	1.24	1.8%	69.6	65.8	8	8	100.0%
31 Mar 1990	68.0	98.5%	.78	1.2%	69.1	66.7	8	8	100.0%
1 Apr 1990	68.8	99.7%	.84	1.2%	69.7	67.7	6	6	100.0%
2 Apr 1990	68.1	98.7%	.62	.9%	69.2	67.4	6	6	100.0%
3 Apr 1990	67.6	98.0%	.62	.9%	68.7	66.9	8	8	100.0%
4 Apr 1990	67.8	98.2%	.68	1.0%	68.7	66.7	6	6	100.0%
5 Apr 1990	68.3	99.1%	.49	.7%	69.2	67.8	6	6	100.0%
6 Apr 1990	68.1	98.7%	1.02	1.5%	69.5	66.7	7	7	100.0%
7 Apr 1990	67.8	98.3%	.71	1.1%	68.9	67.2	7	7	100.0%
8 Apr 1990	67.8	98.2%	.68	1.0%	69.0	67.2	6	6	100.0%
9 Apr 1990	67.8	97.9%	.60	.9%	68.6	66.9	8	8	100.0%
10 Apr 1990	67.9	98.4%	.42	.6%	68.3	67.4	5	5	100.0%
11 Apr 1990	68.9	99.8%	.71	1.0%	69.9	68.1	7	7	100.0%
12 Apr 1990	69.0	100.0%	.64	.9%	69.9	68.2	6	6	100.0%
13 Apr 1990	68.8	99.7%	.54	.8%	69.3	68.1	8	8	100.0%
14 Apr 1990	68.9	99.9%	.30	.4%	69.5	68.6	7	7	100.0%
15 Apr 1990	68.9	99.9%	.37	.5%	69.4	68.6	7	7	100.0%
16 Apr 1990	69.1	100.2%	.44	.6%	69.7	68.5	7	7	100.0%
17 Apr 1990	69.5	100.7%	.43	.6%	70.1	68.9	7	7	100.0%
18 Apr 1990	69.5	100.7%	.59	.8%	70.2	68.7	6	6	100.0%
19 Apr 1990	67.5	97.9%	.56	.8%	68.3	66.7	7	7	100.0%
Summary	68.2	98.9%	.95	1.4%	70.2	65.7	161	161	100.0%

Chloroprene Develop. Toxicity (Rabbits)

175 ppm (B)

Mean & Standard Deviation

From 27 Mar 1990 through 19 Apr 1990

Relative Humidity Data

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 0 ppm (A)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	52.0	94.5%	3.10	6.0%	56.0	47.0	6	6	100.0%
28 Mar 1990	58.9	107.0%	5.84	9.9%	67.0	52.0	8	8	100.0%
29 Mar 1990	58.8	106.8%	4.83	8.2%	64.0	52.0	8	8	100.0%
30 Mar 1990	61.1	111.1%	5.51	9.0%	67.0	52.0	8	8	100.0%
31 Mar 1990	62.3	113.2%	5.41	8.7%	68.0	55.0	7	7	100.0%
1 Apr 1990	59.8	108.6%	5.63	9.4%	66.0	52.0	8	8	100.0%
2 Apr 1990	63.1	114.8%	4.10	6.5%	68.0	58.0	7	7	100.0%
3 Apr 1990	61.5	111.8%	5.13	8.3%	68.0	55.0	8	8	100.0%
4 Apr 1990	59.5	108.2%	5.71	9.6%	68.0	52.0	8	8	100.0%
5 Apr 1990	59.6	108.4%	5.97	10.0%	68.0	53.0	8	8	100.0%
6 Apr 1990	62.5	113.6%	4.84	7.7%	68.0	56.0	8	8	100.0%
7 Apr 1990	60.4	109.8%	4.87	8.1%	68.0	55.0	8	8	100.0%
8 Apr 1990	56.3	102.3%	8.91	15.8%	69.0	47.0	8	8	100.0%
9 Apr 1990	60.0	109.1%	4.76	7.9%	66.0	52.0	7	7	100.0%
10 Apr 1990	57.6	104.7%	5.47	9.5%	64.0	51.0	7	7	100.0%
11 Apr 1990	58.9	107.0%	4.12	7.0%	64.0	54.0	8	8	100.0%
12 Apr 1990	55.6	101.1%	4.63	8.3%	61.0	48.0	8	8	100.0%
13 Apr 1990	60.9	110.6%	4.60	7.6%	66.0	52.0	7	7	100.0%
14 Apr 1990	61.3	111.4%	2.98	4.9%	67.0	58.0	7	7	100.0%
15 Apr 1990	57.4	104.4%	1.51	2.6%	60.0	56.0	7	7	100.0%
16 Apr 1990	59.5	108.2%	3.85	6.5%	64.0	54.0	8	8	100.0%
17 Apr 1990	55.6	101.1%	5.10	9.2%	63.0	49.0	8	8	100.0%
18 Apr 1990	52.1	94.8%	2.30	4.4%	55.0	49.0	8	8	100.0%
19 Apr 1990	53.7	97.7%	5.09	9.5%	61.0	46.0	8	8	100.0%
Summary	58.7	106.7%	5.55	9.5%	69.0	46.0	183	183	100.0%

Chloroprene Develop. Toxicity (Rabbits)

0 ppm (A)

Mean & Standard Deviation

From 27 Mar 1990 through 19 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 0 ppm (B)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	65.8	119.7%	6.62	10.0%	75.0	60.0	6	4	66.7%
24 Mar 1990	51.6	93.9%	4.69	9.1%	63.0	49.0	8	8	100.0%
25 Mar 1990	48.6	88.4%	2.97	6.1%	51.0	42.0	8	8	100.0%
26 Mar 1990	53.0	96.4%	6.99	13.2%	62.0	45.0	8	8	100.0%
27 Mar 1990	61.9	112.5%	6.20	10.0%	69.0	52.0	7	7	100.0%
28 Mar 1990	64.2	116.8%	7.11	11.1%	73.0	55.0	8	6	75.0%
29 Mar 1990	63.8	115.9%	4.71	7.4%	70.0	56.0	8	8	100.0%
30 Mar 1990	64.7	117.7%	5.85	9.0%	72.0	56.0	8	7	87.5%
31 Mar 1990	67.3	122.3%	4.19	6.2%	71.0	61.0	7	4	57.1%
1 Apr 1990	66.7	121.4%	4.74	7.1%	73.0	60.0	8	6	75.0%
2 Apr 1990	65.2	118.5%	4.45	6.8%	71.0	60.0	6	5	83.3%
3 Apr 1990	65.9	119.8%	3.23	4.9%	69.0	60.0	8	8	100.0%
4 Apr 1990	62.0	112.7%	5.35	8.6%	70.0	54.0	8	8	100.0%
5 Apr 1990	63.4	115.2%	4.03	6.4%	69.0	58.0	8	8	100.0%
6 Apr 1990	62.9	114.3%	5.46	8.7%	69.0	56.0	7	7	100.0%
7 Apr 1990	64.4	117.0%	3.85	6.0%	69.0	59.0	8	8	100.0%
8 Apr 1990	58.8	106.8%	7.80	13.3%	68.0	51.0	8	8	100.0%
9 Apr 1990	64.7	117.7%	5.79	9.0%	70.0	55.0	7	7	100.0%
10 Apr 1990	64.9	117.9%	6.09	9.4%	71.0	56.0	7	5	71.4%
11 Apr 1990	65.6	119.3%	6.50	9.9%	72.0	56.0	8	5	62.5%
12 Apr 1990	62.3	113.2%	5.99	9.6%	70.0	53.0	8	8	100.0%
13 Apr 1990	66.7	121.3%	5.15	7.7%	72.0	59.0	7	4	57.1%
14 Apr 1990	64.4	117.1%	4.04	6.3%	73.0	61.0	7	6	85.7%
15 Apr 1990	61.6	111.9%	2.23	3.6%	64.0	58.0	7	7	100.0%
16 Apr 1990	64.2	116.8%	4.95	7.7%	70.0	56.0	8	8	100.0%
17 Apr 1990	59.6	108.4%	4.50	7.6%	67.0	54.0	8	8	100.0%
18 Apr 1990	56.3	102.3%	2.66	4.7%	59.0	51.0	8	8	100.0%
Summary	62.1	112.9%	6.82	11.0%	75.0	42.0	204	184	90.2%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 10 ppm (A)				/Relative Humidity		Range= 40.0 to 70.0			
Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	68.0	120.0%	7.16	10.8%	75.0	56.0	6	5	83.3%
24 Mar 1990	58.0	105.5%	5.48	9.4%	70.0	53.0	8	8	100.0%
25 Mar 1990	57.1	103.9%	2.95	5.2%	60.0	52.0	8	8	100.0%
26 Mar 1990	59.0	107.3%	8.90	15.1%	69.0	45.0	8	8	100.0%
27 Mar 1990	64.7	117.7%	7.20	11.1%	72.0	55.0	7	5	71.4%
28 Mar 1990	65.1	118.4%	9.37	14.4%	77.0	54.0	8	4	50.0%
29 Mar 1990	55.2	100.5%	9.45	17.1%	66.0	43.0	8	8	100.0%
30 Mar 1990	57.3	104.1%	8.28	14.5%	64.0	43.0	8	8	100.0%
31 Mar 1990	57.6	104.7%	7.76	13.5%	65.0	47.0	7	7	100.0%
1 Apr 1990	58.4	106.1%	7.61	13.0%	67.0	46.0	8	8	100.0%
2 Apr 1990	58.6	102.9%	6.24	11.0%	64.0	50.0	7	7	100.0%
3 Apr 1990	60.1	109.3%	6.51	10.8%	70.0	49.0	8	8	100.0%
4 Apr 1990	60.1	109.3%	9.28	15.4%	72.0	48.0	8	7	87.5%
5 Apr 1990	58.5	106.4%	9.13	15.6%	70.0	48.0	8	8	100.0%
6 Apr 1990	61.3	111.4%	11.13	18.2%	74.0	48.0	7	4	57.1%
7 Apr 1990	61.6	112.0%	9.35	15.2%	72.0	48.0	8	7	87.5%
8 Apr 1990	58.1	105.7%	11.61	20.0%	74.0	46.0	8	6	75.0%
9 Apr 1990	67.1	122.1%	8.86	13.2%	75.0	54.0	7	3	42.9%
10 Apr 1990	59.4	108.1%	9.95	16.7%	76.0	48.0	7	6	85.7%
11 Apr 1990	57.5	104.5%	5.78	10.1%	63.0	50.0	8	8	100.0%
12 Apr 1990	54.5	99.1%	6.80	12.5%	62.0	43.0	8	8	100.0%
13 Apr 1990	58.0	105.5%	6.08	10.5%	65.0	47.0	7	7	100.0%
14 Apr 1990	59.0	107.3%	4.20	7.1%	68.0	55.0	7	7	100.0%
15 Apr 1990	53.6	97.4%	2.94	5.5%	58.0	49.0	7	7	100.0%
16 Apr 1990	55.9	101.6%	6.10	10.9%	64.0	46.0	8	8	100.0%
17 Apr 1990	54.0	98.2%	4.44	8.2%	62.0	48.0	8	8	100.0%
18 Apr 1990	50.8	92.3%	5.16	10.2%	60.0	43.0	8	8	100.0%
Summary	58.6	106.5%	8.08	13.8%	77.0	43.0	205	186	90.7%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 10 ppm (B)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	59.0	107.3%	3.85	6.5%	64.0	54.0	8	8	100.0%
28 Mar 1990	62.0	112.7%	6.46	10.4%	71.0	52.0	8	7	87.5%
29 Mar 1990	53.6	97.5%	6.28	11.7%	65.0	46.0	8	8	100.0%
30 Mar 1990	55.7	101.4%	6.50	11.7%	63.0	45.0	8	8	100.0%
31 Mar 1990	55.4	100.8%	5.83	10.5%	62.0	46.0	7	7	100.0%
1 Apr 1990	59.1	107.5%	7.04	11.9%	66.0	49.0	8	8	100.0%
2 Apr 1990	57.9	105.2%	4.74	8.2%	63.0	52.0	7	7	100.0%
3 Apr 1990	59.3	107.7%	5.23	8.8%	65.0	51.0	8	8	100.0%
4 Apr 1990	54.7	99.5%	7.81	14.3%	63.0	44.0	8	8	100.0%
5 Apr 1990	55.6	101.1%	8.03	14.4%	65.0	46.0	8	8	100.0%
6 Apr 1990	56.6	102.9%	6.24	11.0%	65.0	49.0	7	7	100.0%
7 Apr 1990	56.0	101.8%	6.97	12.4%	64.0	46.0	8	8	100.0%
8 Apr 1990	53.5	97.3%	7.73	14.4%	65.0	46.0	8	8	100.0%
9 Apr 1990	57.0	103.6%	5.86	10.3%	62.0	48.0	7	7	100.0%
10 Apr 1990	57.6	104.7%	5.22	9.1%	64.0	50.0	7	7	100.0%
11 Apr 1990	60.0	109.1%	6.74	11.2%	69.0	52.0	8	8	100.0%
12 Apr 1990	57.0	103.6%	7.15	12.5%	66.0	45.0	8	8	100.0%
13 Apr 1990	56.0	101.8%	4.86	8.7%	66.0	50.0	7	7	100.0%
14 Apr 1990	60.1	109.4%	2.27	3.8%	62.0	56.0	7	7	100.0%
15 Apr 1990	57.6	104.7%	1.27	2.2%	59.0	56.0	7	7	100.0%
16 Apr 1990	60.5	110.0%	6.12	10.1%	66.0	50.0	8	8	100.0%
17 Apr 1990	54.0	98.2%	5.90	10.9%	65.0	45.0	8	8	100.0%
18 Apr 1990	49.1	89.3%	2.75	5.6%	53.0	45.0	8	8	100.0%
19 Apr 1990	56.1	102.0%	4.58	8.2%	63.0	50.0	8	8	100.0%
Summary	56.8	103.2%	6.23	11.0%	71.0	44.0	182	181	99.5%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 40 ppm (A)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	53.7	97.6%	6.06	11.3%	60.0	46.0	8	8	100.0%
24 Mar 1990	47.1	85.7%	6.13	13.0%	60.0	40.0	8	8	100.0%
25 Mar 1990	47.1	85.7%	4.49	9.5%	51.0	39.0	8	7	87.5%
26 Mar 1990	60.8	110.5%	11.85	19.2%	73.0	38.0	8	5	62.5%
27 Mar 1990	68.4	124.4%	7.00	10.2%	75.0	58.0	7	3	42.9%
28 Mar 1990	64.9	118.0%	7.90	12.2%	74.0	54.0	8	6	75.0%
29 Mar 1990	54.2	98.6%	8.03	14.8%	68.0	45.0	8	8	100.0%
30 Mar 1990	56.9	103.4%	7.85	13.8%	65.0	44.0	8	8	100.0%
31 Mar 1990	58.6	106.5%	7.63	13.0%	66.0	48.0	7	7	100.0%
1 Apr 1990	59.3	107.7%	7.17	12.1%	67.0	50.0	8	8	100.0%
2 Apr 1990	58.3	106.1%	6.47	11.1%	64.0	50.0	6	6	100.0%
3 Apr 1990	59.5	108.2%	6.57	11.0%	66.0	49.0	8	8	100.0%
4 Apr 1990	54.1	98.4%	6.89	12.7%	63.0	46.0	7	7	100.0%
5 Apr 1990	56.1	102.0%	7.90	14.1%	65.0	46.0	8	8	100.0%
6 Apr 1990	60.7	110.4%	7.43	12.2%	68.0	50.0	7	7	100.0%
7 Apr 1990	59.8	108.6%	6.48	10.8%	68.0	50.0	8	8	100.0%
8 Apr 1990	54.7	99.5%	6.92	12.6%	63.0	47.0	8	8	100.0%
9 Apr 1990	58.4	106.2%	6.50	11.1%	63.0	49.0	7	7	100.0%
10 Apr 1990	55.6	101.0%	5.32	9.6%	64.0	49.0	7	7	100.0%
11 Apr 1990	57.5	104.5%	6.35	11.0%	66.0	50.0	8	8	100.0%
12 Apr 1990	52.7	95.9%	7.38	14.0%	61.0	40.0	8	8	100.0%
13 Apr 1990	51.1	93.0%	4.88	9.5%	60.0	45.0	7	7	100.0%
14 Apr 1990	56.1	102.1%	3.58	6.4%	59.0	49.0	7	7	100.0%
15 Apr 1990	54.0	98.2%	2.77	5.1%	56.0	50.0	7	7	100.0%
16 Apr 1990	56.3	102.3%	5.15	9.2%	61.0	48.0	7	7	100.0%
17 Apr 1990	53.6	97.5%	4.75	8.9%	62.0	46.0	8	8	100.0%
18 Apr 1990	48.5	88.2%	1.11	0.5%	54.0	43.0	8	8	100.0%
Summary	56.2	102.1%	7.87	14.0%	75.0	38.0	202	192	95.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 40 ppm (B)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	64.3	117.0%	3.27	5.1%	68.0	59.0	6	6	100.0%
28 Mar 1990	68.8	125.0%	5.63	8.2%	76.0	61.0	8	4	50.0%
29 Mar 1990	56.9	103.4%	4.94	8.7%	62.0	48.0	8	8	100.0%
30 Mar 1990	59.6	108.4%	8.31	13.9%	69.0	47.0	8	8	100.0%
31 Mar 1990	63.0	114.5%	6.48	10.3%	70.0	53.0	7	7	100.0%
1 Apr 1990	62.0	112.7%	7.52	12.1%	69.0	50.0	8	8	100.0%
2 Apr 1990	60.7	110.3%	5.82	9.6%	67.0	53.0	6	6	100.0%
3 Apr 1990	60.5	110.0%	6.39	10.6%	66.0	51.0	8	8	100.0%
4 Apr 1990	54.4	99.0%	7.14	13.1%	65.0	44.0	7	7	100.0%
5 Apr 1990	56.1	102.0%	6.24	11.1%	63.0	48.0	8	8	100.0%
6 Apr 1990	59.1	107.5%	6.62	11.2%	64.0	50.0	7	7	100.0%
7 Apr 1990	57.8	105.0%	5.87	10.2%	63.0	48.0	8	8	100.0%
8 Apr 1990	54.0	98.2%	7.67	14.2%	65.0	47.0	8	8	100.0%
9 Apr 1990	60.4	109.9%	5.88	9.7%	65.0	51.0	7	7	100.0%
10 Apr 1990	57.3	104.2%	5.44	9.5%	63.0	49.0	7	7	100.0%
11 Apr 1990	58.3	105.9%	5.70	9.8%	67.0	52.0	8	8	100.0%
12 Apr 1990	57.9	105.2%	7.14	12.3%	66.0	47.0	8	8	100.0%
13 Apr 1990	61.9	112.5%	4.85	7.8%	66.0	52.0	7	7	100.0%
14 Apr 1990	59.6	108.3%	2.82	4.7%	65.0	56.0	7	7	100.0%
15 Apr 1990	56.9	103.4%	1.68	2.9%	59.0	55.0	7	7	100.0%
16 Apr 1990	60.1	109.4%	4.22	7.0%	65.0	54.0	7	7	100.0%
17 Apr 1990	55.6	101.1%	3.78	6.8%	62.0	52.0	8	8	100.0%
18 Apr 1990	52.6	95.7%	3.11	5.9%	59.0	48.0	8	8	100.0%
19 Apr 1990	56.0	101.8%	2.98	5.3%	61.0	53.0	8	8	100.0%
Summary	58.8	106.9%	6.40	10.9%	76.0	44.0	179	175	97.8%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 175 ppm (A)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	53.7	97.6%	6.98	13.0%	62.0	45.0	6	6	100.0%
24 Mar 1990	49.0	89.1%	4.31	8.8%	58.0	44.0	8	8	100.0%
25 Mar 1990	47.1	85.7%	3.44	7.3%	51.0	40.0	8	8	100.0%
26 Mar 1990	53.0	96.4%	6.51	12.3%	60.0	40.0	7	7	100.0%
27 Mar 1990	60.6	110.1%	6.73	11.1%	68.0	52.0	7	7	100.0%
28 Mar 1990	63.4	115.3%	6.11	9.6%	71.0	55.0	7	6	85.7%
29 Mar 1990	51.1	93.0%	6.51	12.7%	61.0	42.0	8	8	100.0%
30 Mar 1990	52.9	96.1%	6.41	12.1%	60.0	44.0	7	7	100.0%
31 Mar 1990	55.7	101.3%	6.18	11.1%	62.0	47.0	7	7	100.0%
1 Apr 1990	56.8	103.2%	7.13	12.6%	64.0	46.0	8	8	100.0%
2 Apr 1990	57.7	104.8%	6.31	10.9%	64.0	49.0	6	6	100.0%
3 Apr 1990	58.3	105.9%	6.98	12.0%	64.0	47.0	8	8	100.0%
4 Apr 1990	51.3	93.2%	7.32	14.3%	61.0	41.0	7	7	100.0%
5 Apr 1990	55.0	100.0%	9.07	16.5%	64.0	43.0	7	7	100.0%
6 Apr 1990	55.0	100.0%	6.43	11.7%	62.0	46.0	7	7	100.0%
7 Apr 1990	53.5	97.3%	6.46	12.1%	61.0	45.0	8	8	100.0%
8 Apr 1990	50.1	91.1%	6.85	13.7%	60.0	43.0	8	8	100.0%
9 Apr 1990	55.6	101.0%	7.28	13.1%	64.0	44.0	7	7	100.0%
10 Apr 1990	54.6	99.2%	6.45	11.8%	61.0	42.0	7	7	100.0%
11 Apr 1990	56.8	103.2%	6.07	10.7%	65.0	49.0	8	8	100.0%
12 Apr 1990	60.1	109.3%	7.10	11.8%	67.0	48.0	8	8	100.0%
13 Apr 1990	62.0	112.7%	7.12	11.5%	68.0	49.0	7	7	100.0%
14 Apr 1990	60.3	109.6%	4.11	6.8%	68.0	55.0	7	7	100.0%
15 Apr 1990	57.6	104.7%	1.62	2.8%	59.0	55.0	7	7	100.0%
16 Apr 1990	60.3	109.6%	6.82	11.3%	67.0	51.0	7	7	100.0%
17 Apr 1990	53.9	98.0%	6.98	13.0%	65.0	44.0	8	8	100.0%
18 Apr 1990	52.1	94.8%	6.10	11.7%	63.0	44.0	8	8	100.0%
Summary	55.3	100.6%	7.22	13.0%	71.0	40.0	198	197	99.5%

Chloroprene Develop. Toxicity (Rabbits)

175 ppm (A)

Mean & Standard Deviation

From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 175 ppm (B)

/Relative Humidity

Range= 40.0 to 70.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	56.3	102.4%	6.31	11.2%	62.0	45.0	6	6	100.0%
28 Mar 1990	62.7	114.0%	6.45	10.3%	74.0	57.0	7	6	85.7%
29 Mar 1990	48.1	87.5%	5.08	10.6%	56.0	41.0	8	8	100.0%
30 Mar 1990	52.1	94.8%	8.65	16.6%	61.0	39.0	7	6	85.7%
31 Mar 1990	56.3	102.4%	6.22	11.0%	63.0	48.0	6	6	100.0%
1 Apr 1990	58.5	106.4%	6.80	11.6%	65.0	48.0	8	8	100.0%
2 Apr 1990	58.5	106.4%	5.72	9.8%	65.0	51.0	6	6	100.0%
3 Apr 1990	58.1	105.7%	6.27	10.8%	63.0	49.0	8	8	100.0%
4 Apr 1990	53.9	97.9%	8.15	15.1%	64.0	42.0	7	7	100.0%
5 Apr 1990	56.7	103.1%	5.99	10.6%	63.0	48.0	7	7	100.0%
6 Apr 1990	56.5	102.7%	6.53	11.6%	64.0	49.0	6	6	100.0%
7 Apr 1990	53.7	97.7%	7.03	13.1%	60.0	42.0	8	8	100.0%
8 Apr 1990	49.2	89.5%	6.94	14.1%	60.0	43.0	8	8	100.0%
9 Apr 1990	52.4	95.3%	6.24	11.9%	60.0	43.0	7	7	100.0%
10 Apr 1990	52.6	95.6%	5.80	11.0%	58.0	41.0	7	7	100.0%
11 Apr 1990	54.0	98.2%	5.32	9.8%	62.0	47.0	8	8	100.0%
12 Apr 1990	50.3	91.4%	5.50	10.9%	56.0	40.0	7	7	100.0%
13 Apr 1990	55.3	100.5%	6.63	12.0%	63.0	44.0	7	7	100.0%
14 Apr 1990	56.7	103.1%	2.29	4.0%	61.0	54.0	7	7	100.0%
15 Apr 1990	54.9	99.7%	2.12	3.9%	57.0	51.0	7	7	100.0%
16 Apr 1990	52.9	96.1%	4.30	8.1%	56.0	45.0	7	7	100.0%
17 Apr 1990	50.4	91.6%	4.72	9.4%	56.0	43.0	8	8	100.0%
18 Apr 1990	48.7	88.6%	3.96	8.1%	53.0	43.0	8	8	100.0%
19 Apr 1990	53.4	97.0%	4.90	9.2%	60.0	44.0	8	8	100.0%
Summary	54.1	98.4%	6.53	12.1%	74.0	39.0	173	171	98.8%

Exhaust Airflow Data

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 0 ppm (A)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	14.5	96.8%	.10	.7%	14.7	14.3	7	7	100.0%
28 Mar 1990	15.0	100.0%	.28	1.9%	15.2	14.5	8	8	100.0%
29 Mar 1990	15.0	100.0%	.10	.6%	15.2	14.9	7	7	100.0%
30 Mar 1990	14.9	99.0%	.06	.4%	15.0	14.8	8	8	100.0%
31 Mar 1990	14.8	98.4%	.07	.4%	14.8	14.7	8	8	100.0%
1 Apr 1990	14.7	98.2%	.05	.4%	14.8	14.7	8	8	100.0%
2 Apr 1990	14.6	97.2%	.03	.2%	14.6	14.5	7	7	100.0%
3 Apr 1990	14.5	96.7%	.03	.2%	14.6	14.5	7	7	100.0%
4 Apr 1990	14.3	95.6%	.05	.4%	14.4	14.3	7	7	100.0%
5 Apr 1990	14.1	94.2%	.08	.5%	14.3	14.1	7	7	100.0%
6 Apr 1990	14.5	97.0%	.42	2.9%	14.9	14.1	7	7	100.0%
7 Apr 1990	14.8	98.8%	.07	.5%	15.0	14.7	8	8	100.0%
8 Apr 1990	14.7	98.1%	.12	.8%	14.9	14.6	7	7	100.0%
9 Apr 1990	14.4	96.2%	.13	.9%	14.6	14.2	8	8	100.0%
10 Apr 1990	14.0	93.5%	.14	1.0%	14.3	13.9	6	6	100.0%
11 Apr 1990	14.6	97.1%	.52	3.6%	15.0	13.9	8	8	100.0%
12 Apr 1990	14.7	98.2%	.11	.8%	14.9	14.6	7	7	100.0%
13 Apr 1990	14.4	96.2%	.16	1.1%	14.6	14.2	7	7	100.0%
14 Apr 1990	15.1	100.7%	.37	2.5%	15.4	14.3	7	7	100.0%
15 Apr 1990	15.2	101.5%	.10	.6%	15.4	15.2	7	7	100.0%
16 Apr 1990	14.8	98.7%	.18	1.2%	15.1	14.7	7	7	100.0%
17 Apr 1990	14.6	97.6%	.13	.9%	14.8	14.5	7	7	100.0%
18 Apr 1990	14.6	97.2%	.76	5.2%	15.4	13.2	7	7	100.0%
19 Apr 1990	15.4	103.0%	.07	.4%	15.5	15.3	8	8	100.0%
Summary	14.7	98.0%	.39	2.6%	15.5	13.2	175	175	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 0 ppm (B)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	15.2	101.2%	.06	.4%	15.2	15.1	7	7	100.0%
24 Mar 1990	15.1	100.9%	.07	.5%	15.2	15.0	8	8	100.0%
25 Mar 1990	15.1	100.4%	.08	.5%	15.2	15.0	8	8	100.0%
26 Mar 1990	15.0	100.1%	.08	.5%	15.1	14.9	7	7	100.0%
27 Mar 1990	15.2	101.4%	.87	5.7%	17.4	14.8	8	8	100.0%
28 Mar 1990	14.8	98.8%	.06	.4%	14.9	14.7	8	8	100.0%
29 Mar 1990	14.7	98.3%	.07	.4%	14.9	14.7	7	7	100.0%
30 Mar 1990	14.7	97.7%	.03	.2%	14.7	14.6	8	8	100.0%
31 Mar 1990	14.7	97.7%	.02	.1%	14.7	14.6	8	8	100.0%
1 Apr 1990	14.6	97.5%	.01	.1%	14.6	14.6	8	8	100.0%
2 Apr 1990	14.6	97.2%	.04	.3%	14.7	14.5	7	7	100.0%
3 Apr 1990	14.5	96.9%	.02	.2%	14.6	14.5	7	7	100.0%
4 Apr 1990	14.5	96.6%	.03	.2%	14.5	14.5	7	7	100.0%
5 Apr 1990	14.5	96.4%	.04	.3%	14.5	14.4	7	7	100.0%
6 Apr 1990	14.5	96.4%	.03	.2%	14.5	14.4	7	7	100.0%
7 Apr 1990	14.5	96.4%	.03	.2%	14.5	14.4	8	8	100.0%
8 Apr 1990	14.4	96.2%	.04	.3%	14.5	14.4	7	7	100.0%
9 Apr 1990	14.4	95.8%	.05	.4%	14.5	14.3	8	8	100.0%
10 Apr 1990	14.3	95.5%	.03	.2%	14.4	14.3	6	6	100.0%
11 Apr 1990	14.8	98.8%	.43	2.9%	15.1	14.3	8	8	100.0%
12 Apr 1990	15.1	100.7%	.04	.3%	15.2	15.1	7	7	100.0%
13 Apr 1990	15.0	100.2%	.06	.4%	15.1	14.9	7	7	100.0%
14 Apr 1990	15.3	102.1%	.16	1.0%	15.4	15.0	7	7	100.0%
15 Apr 1990	15.4	102.5%	.04	.2%	15.4	15.3	7	7	100.0%
16 Apr 1990	15.4	102.3%	.05	.3%	15.4	15.3	7	7	100.0%
17 Apr 1990	15.3	101.7%	.05	.3%	15.3	15.2	7	7	100.0%
18 Apr 1990	15.2	101.1%	.79	5.2%	15.9	13.5	7	7	100.0%
Summary	14.8	98.9%	.41	2.7%	17.4	13.5	198	198	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 10 ppm (A)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	14.8	98.8%	.08	.5%	15.0	14.7	7	7	100.0%
24 Mar 1990	14.7	98.1%	.09	.6%	14.8	14.6	8	8	100.0%
25 Mar 1990	14.6	97.6%	.06	.4%	14.7	14.6	8	8	100.0%
26 Mar 1990	14.7	97.8%	.10	.7%	14.9	14.6	7	7	100.0%
27 Mar 1990	14.6	97.4%	.15	1.0%	14.8	14.4	8	8	100.0%
28 Mar 1990	14.4	95.8%	.15	1.0%	14.5	14.1	8	8	100.0%
29 Mar 1990	14.5	96.7%	.12	.9%	14.7	14.4	7	7	100.0%
30 Mar 1990	14.9	99.3%	.22	1.5%	15.1	14.4	8	8	100.0%
31 Mar 1990	14.9	99.6%	.11	.8%	15.1	14.8	8	8	100.0%
1 Apr 1990	14.9	99.5%	.11	.7%	15.1	14.8	8	8	100.0%
2 Apr 1990	14.9	99.3%	.11	.7%	15.1	14.8	7	7	100.0%
3 Apr 1990	15.0	100.0%	.15	1.0%	15.2	14.9	7	7	100.0%
4 Apr 1990	15.0	100.1%	.14	.9%	15.2	14.9	7	7	100.0%
5 Apr 1990	14.9	99.2%	.11	.7%	15.1	14.8	7	7	100.0%
6 Apr 1990	14.8	98.9%	.12	.8%	15.1	14.8	7	7	100.0%
7 Apr 1990	14.8	98.5%	.12	.8%	15.0	14.7	8	8	100.0%
8 Apr 1990	14.8	98.6%	.12	.8%	15.0	14.6	7	7	100.0%
9 Apr 1990	14.8	99.0%	.13	.9%	15.1	14.7	8	8	100.0%
10 Apr 1990	14.8	98.5%	.11	.7%	15.0	14.7	6	6	100.0%
11 Apr 1990	14.8	98.6%	.15	1.0%	15.0	14.5	8	8	100.0%
12 Apr 1990	14.8	98.7%	.12	.8%	15.0	14.7	7	7	100.0%
13 Apr 1990	14.8	98.5%	.17	1.2%	15.0	14.5	7	7	100.0%
14 Apr 1990	14.8	99.0%	.13	.9%	15.1	14.7	7	7	100.0%
15 Apr 1990	14.8	98.5%	.13	.9%	15.0	14.7	7	7	100.0%
16 Apr 1990	15.2	101.1%	.17	1.1%	15.4	15.0	7	7	100.0%
17 Apr 1990	15.0	100.3%	.35	2.3%	15.8	14.8	7	7	100.0%
18 Apr 1990	14.8	98.6%	.72	4.8%	15.2	13.2	7	7	100.0%
Summary	14.8	98.7%	.24	1.6%	15.8	13.2	198	198	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 10 ppm (B)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	14.9	99.3%	.15	1.0%	15.1	14.7	7	7	100.0%
28 Mar 1990	14.7	97.7%	.09	.6%	14.7	14.5	8	8	100.0%
29 Mar 1990	14.7	97.9%	.12	.8%	14.9	14.6	7	7	100.0%
30 Mar 1990	15.0	100.1%	.19	1.3%	15.2	14.6	8	8	100.0%
31 Mar 1990	14.9	99.5%	.12	.8%	15.1	14.8	8	8	100.0%
1 Apr 1990	14.8	98.9%	.24	1.6%	15.0	14.3	8	8	100.0%
2 Apr 1990	14.8	98.6%	.53	3.6%	15.2	13.6	7	7	100.0%
3 Apr 1990	15.0	100.3%	.15	1.0%	15.3	14.9	7	7	100.0%
4 Apr 1990	15.0	100.2%	.15	1.0%	15.3	14.9	7	7	100.0%
5 Apr 1990	14.9	99.4%	.11	.7%	15.1	14.8	7	7	100.0%
6 Apr 1990	14.8	98.9%	.11	.7%	15.1	14.7	7	7	100.0%
7 Apr 1990	14.8	98.5%	.11	.8%	15.0	14.7	8	8	100.0%
8 Apr 1990	14.8	98.6%	.10	.7%	14.9	14.7	7	7	100.0%
9 Apr 1990	14.8	98.9%	.12	.8%	15.0	14.7	8	8	100.0%
10 Apr 1990	14.9	99.6%	.12	.8%	15.2	14.9	6	6	100.0%
11 Apr 1990	14.9	99.2%	.15	1.0%	15.1	14.7	8	8	100.0%
12 Apr 1990	14.9	99.1%	.11	.7%	15.1	14.8	7	7	100.0%
13 Apr 1990	14.9	99.1%	.12	.8%	15.0	14.8	6	6	100.0%
14 Apr 1990	14.8	98.6%	.11	.7%	15.0	14.7	7	7	100.0%
15 Apr 1990	14.7	98.0%	.12	.8%	14.9	14.6	7	7	100.0%
16 Apr 1990	14.6	97.5%	.17	1.1%	14.9	14.4	7	7	100.0%
17 Apr 1990	14.6	97.2%	.10	.7%	14.7	14.5	7	7	100.0%
18 Apr 1990	14.4	96.2%	.79	5.5%	14.9	12.7	7	7	100.0%
19 Apr 1990	14.9	99.2%	.11	.7%	15.0	14.7	8	8	100.0%
Summary	14.8	98.8%	.26	1.7%	15.3	12.7	174	174	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 40 ppm (A)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	15.1	100.8%	.07	.5%	15.2	15.0	7	7	100.0%
24 Mar 1990	15.0	100.0%	.08	.5%	15.1	14.8	8	8	100.0%
25 Mar 1990	14.9	99.6%	.06	.4%	15.0	14.9	8	8	100.0%
26 Mar 1990	14.9	99.4%	.08	.5%	15.1	14.8	7	7	100.0%
27 Mar 1990	14.7	97.7%	.14	1.0%	14.9	14.4	8	8	100.0%
28 Mar 1990	14.5	96.4%	.07	.5%	14.5	14.3	8	8	100.0%
29 Mar 1990	14.6	97.0%	.06	.4%	14.7	14.5	7	7	100.0%
30 Mar 1990	14.8	98.9%	.15	1.0%	15.0	14.5	8	8	100.0%
31 Mar 1990	14.8	98.9%	.05	.4%	14.9	14.8	8	8	100.0%
1 Apr 1990	14.9	99.0%	.04	.2%	14.9	14.8	8	8	100.0%
2 Apr 1990	14.8	98.9%	.05	.4%	14.9	14.8	7	7	100.0%
3 Apr 1990	14.9	99.1%	.10	.7%	15.0	14.8	7	7	100.0%
4 Apr 1990	14.9	99.5%	.09	.6%	15.1	14.8	7	7	100.0%
5 Apr 1990	14.8	99.0%	.09	.6%	15.0	14.7	7	7	100.0%
6 Apr 1990	14.9	99.2%	.08	.5%	15.1	14.8	7	7	100.0%
7 Apr 1990	14.8	98.6%	.09	.6%	14.9	14.7	8	8	100.0%
8 Apr 1990	14.8	98.4%	.09	.6%	14.9	14.6	7	7	100.0%
9 Apr 1990	14.8	98.9%	.06	.4%	14.9	14.7	8	8	100.0%
10 Apr 1990	14.7	98.1%	.08	.5%	14.9	14.7	6	6	100.0%
11 Apr 1990	14.8	98.7%	.13	.9%	15.0	14.6	8	8	100.0%
12 Apr 1990	14.8	98.7%	.06	.4%	14.9	14.8	7	7	100.0%
13 Apr 1990	14.8	98.4%	.11	.8%	14.9	14.6	7	7	100.0%
14 Apr 1990	14.8	98.6%	.09	.6%	15.0	14.7	7	7	100.0%
15 Apr 1990	14.7	97.9%	.07	.5%	14.8	14.6	7	7	100.0%
16 Apr 1990	14.6	97.4%	.10	.7%	14.8	14.5	7	7	100.0%
17 Apr 1990	14.6	97.4%	.08	.5%	14.7	14.6	7	7	100.0%
18 Apr 1990	14.5	97.0%	.66	4.6%	14.9	13.1	7	7	100.0%
Summary	14.8	98.6%	.20	1.4%	15.2	13.1	198	198	100.0%

Chloroprene Develop. Toxicity (Rabbits)

40 ppm (A)

Mean & Standard Deviation

From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 40 ppm (B)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	15.0	100.0%	.13	.8%	15.2	14.8	7	7	100.0%
28 Mar 1990	14.8	98.6%	.11	.7%	14.9	14.6	8	8	100.0%
29 Mar 1990	14.8	98.6%	.13	.9%	15.0	14.7	7	7	100.0%
30 Mar 1990	15.1	100.9%	.21	1.4%	15.4	14.7	8	8	100.0%
31 Mar 1990	15.1	100.9%	.08	.6%	15.3	15.0	8	8	100.0%
1 Apr 1990	15.2	101.0%	.08	.5%	15.3	15.0	8	8	100.0%
2 Apr 1990	15.2	101.1%	.08	.5%	15.3	15.1	7	7	100.0%
3 Apr 1990	15.2	101.4%	.12	.8%	15.3	15.1	7	7	100.0%
4 Apr 1990	15.4	102.7%	.12	.8%	15.6	15.3	7	7	100.0%
5 Apr 1990	15.3	101.8%	.11	.7%	15.5	15.2	7	7	100.0%
6 Apr 1990	15.2	101.4%	.10	.6%	15.4	15.1	7	7	100.0%
7 Apr 1990	15.1	100.9%	.10	.7%	15.3	15.1	8	8	100.0%
8 Apr 1990	15.1	100.9%	.10	.7%	15.3	15.0	7	7	100.0%
9 Apr 1990	15.2	101.4%	.13	.8%	15.4	15.0	8	8	100.0%
10 Apr 1990	15.2	101.2%	.09	.6%	15.3	15.1	6	6	100.0%
11 Apr 1990	15.5	103.1%	.19	1.2%	15.7	15.2	8	8	100.0%
12 Apr 1990	15.1	100.9%	.36	2.4%	15.7	14.9	7	7	100.0%
13 Apr 1990	14.9	99.3%	.14	.9%	15.1	14.7	7	7	100.0%
14 Apr 1990	14.9	99.2%	.09	.6%	15.0	14.8	7	7	100.0%
15 Apr 1990	14.8	98.7%	.11	.7%	15.0	14.7	7	7	100.0%
16 Apr 1990	14.7	98.1%	.13	.9%	14.9	14.6	7	7	100.0%
17 Apr 1990	14.7	97.9%	.09	.6%	14.8	14.6	7	7	100.0%
18 Apr 1990	14.6	97.3%	.66	4.5%	15.0	13.1	7	7	100.0%
19 Apr 1990	14.9	99.6%	.09	.6%	15.1	14.6	8	8	100.0%
Summary	15.0	100.3%	.28	1.9%	15.7	13.1	175	175	100.0%

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 23 Mar 1990 through 18 Apr 1990

Summary Data for: 175 ppm (A)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
23 Mar 1990	14.9	99.0%	.08	.5%	15.0	14.7	7	7	100.0%
24 Mar 1990	14.8	98.4%	.08	.6%	14.9	14.6	8	8	100.0%
25 Mar 1990	14.7	98.0%	.07	.5%	14.8	14.6	8	8	100.0%
26 Mar 1990	14.7	98.1%	.09	.6%	14.9	14.6	7	7	100.0%
27 Mar 1990	14.7	98.2%	.14	1.0%	15.0	14.6	8	8	100.0%
28 Mar 1990	14.6	97.2%	.13	.9%	14.7	14.3	8	8	100.0%
29 Mar 1990	14.7	98.1%	.10	.7%	14.9	14.6	7	7	100.0%
30 Mar 1990	15.1	100.9%	.23	1.5%	15.4	14.6	8	8	100.0%
31 Mar 1990	15.2	101.0%	.10	.7%	15.3	15.1	8	8	100.0%
1 Apr 1990	15.2	101.1%	.11	.7%	15.3	15.1	8	8	100.0%
2 Apr 1990	15.1	100.8%	.10	.7%	15.3	15.1	7	7	100.0%
3 Apr 1990	15.2	101.0%	.15	1.0%	15.3	15.0	7	7	100.0%
4 Apr 1990	15.1	100.7%	.14	.9%	15.3	15.0	7	7	100.0%
5 Apr 1990	15.2	101.2%	.11	.7%	15.3	15.1	7	7	100.0%
6 Apr 1990	15.3	101.9%	.12	.8%	15.5	15.2	7	7	100.0%
7 Apr 1990	15.3	101.7%	.12	.8%	15.5	15.1	8	8	100.0%
8 Apr 1990	15.3	101.9%	.11	.7%	15.5	15.1	7	7	100.0%
9 Apr 1990	15.3	102.3%	.12	.8%	15.6	15.2	8	8	100.0%
10 Apr 1990	15.3	101.7%	.11	.7%	15.5	15.2	6	6	100.0%
11 Apr 1990	15.2	101.4%	.15	1.0%	15.4	15.0	8	8	100.0%
12 Apr 1990	15.5	103.1%	.15	1.0%	15.7	15.4	7	7	100.0%
13 Apr 1990	15.4	102.9%	.15	1.0%	15.6	15.1	7	7	100.0%
14 Apr 1990	15.0	100.0%	.18	1.2%	15.4	14.9	7	7	100.0%
15 Apr 1990	14.8	98.7%	.11	.8%	15.0	14.7	7	7	100.0%
16 Apr 1990	14.7	98.0%	.19	1.3%	15.0	14.6	7	7	100.0%
17 Apr 1990	14.8	98.7%	.09	.6%	14.9	14.7	7	7	100.0%
18 Apr 1990	14.7	97.9%	.70	4.8%	15.0	13.1	7	7	100.0%
Summary	15.0	100.1%	.31	2.1%	15.7	13.1	198	198	100.0%

Chloroprene Develop. Toxicity (Rabbits)

175 ppm (A)

Mean & Standard Deviation

From 23 Mar 1990 through 18 Apr 1990

Daily Summation For Chloroprene Develop. Toxicity (Rabbits)

From 27 Mar 1990 through 19 Apr 1990

Summary Data for: 175 ppm (B)

/Exhaust Air Flow

Range= 12.0 to 18.0

Date	Mean	% Target	Std Dev	% RSD	Maximum	Minimum	N	N in	% N in
27 Mar 1990	15.3	102.3%	.21	1.3%	15.7	15.1	7	7	100.0%
28 Mar 1990	15.2	101.2%	.15	1.0%	15.3	15.0	8	8	100.0%
29 Mar 1990	15.2	101.7%	.17	1.1%	15.5	15.1	7	7	100.0%
30 Mar 1990	15.2	101.2%	.15	1.0%	15.4	15.1	8	8	100.0%
31 Mar 1990	15.1	100.9%	.11	.7%	15.3	15.0	8	8	100.0%
1 Apr 1990	15.2	101.2%	.13	.8%	15.3	15.1	8	8	100.0%
2 Apr 1990	15.0	100.0%	.13	.9%	15.2	14.9	7	7	100.0%
3 Apr 1990	15.0	100.3%	.17	1.1%	15.3	14.9	7	7	100.0%
4 Apr 1990	15.1	100.5%	.18	1.2%	15.3	15.0	7	7	100.0%
5 Apr 1990	15.4	102.4%	.09	.6%	15.5	15.3	7	7	100.0%
6 Apr 1990	15.3	102.0%	.14	.9%	15.6	15.2	7	7	100.0%
7 Apr 1990	15.2	101.2%	.17	1.1%	15.5	15.1	8	8	100.0%
8 Apr 1990	15.2	101.1%	.15	1.0%	15.4	15.0	7	7	100.0%
9 Apr 1990	15.0	99.9%	.16	1.1%	15.2	14.8	8	8	100.0%
10 Apr 1990	14.9	99.4%	.12	.8%	15.1	14.8	6	6	100.0%
11 Apr 1990	14.7	98.0%	.19	1.3%	14.9	14.4	8	8	100.0%
12 Apr 1990	15.0	100.0%	.15	1.0%	15.3	14.9	7	7	100.0%
13 Apr 1990	14.9	99.5%	.19	1.3%	15.2	14.7	7	7	100.0%
14 Apr 1990	14.8	97.6%	.15	1.0%	14.8	14.5	7	7	100.0%
15 Apr 1990	14.8	97.0%	.16	1.1%	14.8	14.4	7	7	100.0%
16 Apr 1990	14.9	99.7%	.19	1.2%	15.2	14.6	7	7	100.0%
17 Apr 1990	15.0	99.9%	.15	1.0%	15.2	14.9	7	7	100.0%
18 Apr 1990	14.9	99.5%	.74	4.9%	15.4	13.3	7	7	100.0%
19 Apr 1990	15.3	102.1%	.14	.9%	15.5	15.1	8	8	100.0%
Summary	15.1	100.4%	.29	1.9%	15.7	13.3	175	175	100.0%

Exposure Operation Discussion Sheets

EXPOSURE OPERATION DISCUSSION SHEET

INCLUDES DISCUSSIONS AND/OR EXPLANATIONS OF PROBLEMS AFFECTING ANIMAL ENVIRONMENT AND EXPOSURES. EXPLANATIONS ARE INCLUDED FOR DATA IN WHICH THERE WERE EXCURSIONS OF DAILY MEAN OR STANDARD DEVIATION BEYOND ALLOWABLE OPERATING LIMITS OR EXCURSIONS OF INDIVIDUAL DATUM BEYOND CRITICAL LIMITS.

STUDY: IRT Chloroprene Inhalation Teratology Study

REPORTING PERIOD: March 23 - April 19, 1990

NOTE: 24 Hour Collection Period extends from ~5:00 a.m. to ~5:00 a.m.

COMPILED BY: R.J. Weigel *RJW*

DATE: 4/24/90

CHAMBER CONCENTRATION

DATE DISCUSSION OR EXPLANATION

3/29/90 The stream-select valve on the on-line GC failed before the start of the exposure. Exposures were started at the usual time using the purity GC as a monitor. This GC was checked against the standard and agreed. During the day, the exposure operating parameters were kept at the previous day's settings. Four hours into the exposure, the main GC was brought back on-line and subsequent readings agreed with those of the purity GC. Only 4 concentration readings per chamber were obtained during the exposure; no purity data was obtained.

4/6/90 Exposures were terminated after 5 hr 1 min. Chemical consumption in the generator was much higher than normal and it was unlikely that sufficient chemical remained to complete a normal exposure. In addition, small amounts of dimer were picked up by the purity GC. The high consumption was due to a leak in the generator. New seals were put into the generator. A small amount of polymer may also have been on the neck of the flask, preventing a good seal.

TEMPERATURE & RELATIVE HUMIDITY

DATE DISCUSSION OR EXPLANATION

3/23/90 First day in chambers. Temperatures were set low to insure that the animals did not get overheated. However, chamber temperatures did not rise with animal loading as expected. Therefore, the following readings were below the critical low alarm limit (63°F) and the following chamber mean temperatures were below the protocol-defined operating limit of 65°F.

Time	Chamber	Reading	Alarm Limit	Mean
00:56	0 ppm-B	62.9	63	65.1
21:48	10 ppm-A	61.5	63	63.4
00:49		60.7		
04:13		62.2		
21:51	40 ppm-A	62.3	63	63.5
00:53		61.3		
04:16		62.3		

3/24/90 The specialist increased the room temperature early the following morning. Two chambers had temperatures below the critical low alarm limit in the morning before the changes in room temperature were reflected in the chamber temperatures. They were in the 10 ppm-A (62.7°F) and in the 40 ppm-A (62.5°F). Mean temperature in the 40 ppm-A chamber was 64.8°F, below the protocol-defined lower limit of 65°F.

3/28/90 Relative humidity in the 10 ppm A chamber (77% at 00:06) and in the 40 pm B chamber (76% at 03:21) exceeded the critical high alarm limit (75%). Adjustments were made to the wet/dry air mixture.

Battelle
Pacific Northwest Laboratories
Monthly Progress Report
for March - April, 1990

3/29/90 Temperature in the 40 ppm A chamber ranged from 63.7 to 65.4°F. While no temperatures exceeded the critical low alarm limit (63°F), the mean temperature in this chamber was 64.5°F, below the protocol-defined lower operating limit of 65°F.

4/10/90 Relative humidity in the 10 ppm A chamber (76% at 05:36) exceeded the critical high alarm limit (75%). Adjustments were made to the wet/dry air mixture.

CHAMBER FLOW & VACUUM

DATE DISCUSSION OR EXPLANATION

No problems or excursions to report this month.

Chamber Uniformity Data

TPV MEASUREMENTS

TPV MEASUREMENTS												
CHAMBER: 175 ppm B			40 ppm B			10 ppm B						
DATE: 4/5/90			4/4/90			4/3/90						
SAMPLE PORT		MONITOR READING	% of Mean	MONITOR READING		% of Mean	MONITOR READING		% of Mean	MONITOR READING		% of Mean
BACK:		1B										
		2B										
		3B	1671090	99.4%	380639	100.6%	97203	100.0%				
		4B	1674210	99.6%	382356	101.1%	97731	100.6%				
		5B	1683212	100.1%	378682	100.1%	97212	100.0%				
		6B	1685797	100.2%	378113	100.0%	97556	100.4%				
FRONT:		1F										
		2F	1683699	100.1%	379063	100.2%	97265	100.1%				
		3F	1696908	100.9%	379778	100.4%	96207	99.0%				
		4F	1686785	100.3%	376182	99.5%	96998	99.8%				
		5F	1679230	99.9%	373595	98.8%	97246	100.1%				
		6F	1674839	99.6%	375807	99.4%	97135	100.0%				
MEAN:		1681752.2	100.0%	378246.1	100.0%	97172.6	100.0%					
TPV:		7921.73	0.5%	2685.18	0.7%	424.19	0.4%					
BPV:		////////////////	0.4%	////////////////	0.6%	////////////////	0.4%	////////////////		////////////////		

* Indistinguishable from WPV

WPV MEASUREMENTS

IN-LINE 1st	1683699	100.0%	379063	100.3%	97265	100.0%		
2nd	1685335	100.1%	378491	100.2%	97057	99.8%		
3rd	1679556	99.8%	376072	99.5%	97453	100.2%		
MEAN:	1682863.3	100.0%	377875.3	100.0%	97258.3	100.0%		
WPV:	2978.75	0.2%	1587.70	0.4%	198.08	0.2%		

INLET AND EXHAUST MEASUREMENTS

Inlet	1729636	411014	117976		
Exhaust	1633642	375542	97219		
% Difference:	5.5%	8.6%	17.6%		

[Data located in appropriate daily data file]

MONITOR TYPE: HP 5890BNW #: 52642-101

COMMENTS: _____

ENTERED BY: D. FoltzDATE: 4/11/90REVIEWED BY: RJ Weigel *RJW*DATE: 4/19/90

CHAMBER UNIFORMITY DATA SHEET

COMPOUND: ChloropreneEXPOSURE ROOM NUMBER: 432

TPV MEASUREMENTS

CHAMBER:	175 ppm A	40 ppm A	10 ppm A		
DATE:	4/4/90	4/3/90	4/2/90		
SAMPLE PORT	MONITOR READING % of Mean	MONITOR READING % of Mean	MONITOR READING % of Mean	MONITOR READING % of Mean	MONITOR READING % of Mean
BACK: 1B					
2B					
3B	1673970 99.1%	380494 99.5%	94804 100.9%		
4B	1689357 100.0%	385380 100.8%	95801 101.9%		
5B	1685631 99.8%	380447 99.5%	94147 100.2%		
6B	1690149 100.0%	383069 100.2%	94099 100.1%		
FRONT: 1F					
2F	1698376 100.5%	383173 100.2%	94866 100.9%		
3F	1699081 100.6%	380047 99.4%	93699 99.7%		
4F	1685464 99.8%	384416 100.6%	93931 99.9%		
5F	1686928 99.9%	378849 99.1%	91297 97.1%		
6F	1695778 100.4%	384183 100.5%	93368 99.3%		
MEAN:	1689414.9 100.0%	382228.7 100.0%	94001.3 100.0%		
TPV:	7821.63 0.5%	2304.83 0.6%	1248.62 1.3%		
BPV:	////////// 0.4%	////////// 0.6%	////////// 1.0%	//////////	//////////

* Indistinguishable from WPV

WPV MEASUREMENTS

IN-LINE 1st	1698376 100.2%	383173 100.1%	94866 100.5%		
2nd	1695190 100.0%	382156 99.8%	94930 100.5%		
3rd	1691592 99.8%	383021 100.1%	93525 99.0%		
MEAN:	1695052.7 100.0%	382783.3 100.0%	94440.3 100.0%		
WPV:	3394.08 0.2%	548.58 0.1%	793.35 0.8%		

INLET AND EXHAUST MEASUREMENTS

Inlet	1856075	519393	102240		
Exhaust	1677431	378804	93916		
% Difference:	9.6%	27.1%	8.1%		

[Data located in appropriate daily data file]

MONITOR TYPE: HP 5890BNW #: 52642-101COMMENTS: Measurements with animals in chambers.ENTERED BY: D. FoltzDATE: 4/11/90REVIEWED BY: RJ WeigelDATE: 4/19/90

TPV MEASUREMENTS

CHAMBER:	125 B	25 B	5 B		
DATE:	2/20/90	2/17/90	2/17/90		
SAMPLE PORT	MONITOR READING % of Mean	MONITOR READING % of Mean	MONITOR READING % of Mean	MONITOR READING % of Mean	MONITOR READING % of Mean
BACK: 1B	1077512 99.9%	208856 97.2%	43432 100.1%		
2B	1079423 100.0%	213359 99.3%	43486 100.2%		
3B	1078240 99.9%	214873 100.0%	43465 100.1%		
4B	1078768 100.0%	206849 96.3%	43475 100.2%		
5B	1079224 100.0%	216848 101.0%	43509 100.2%		
6B	1080707 100.2%	217240 101.1%	43159 99.4%		
FRONT: 1F	1079578 100.1%	216286 100.7%	43360 99.9%		
2F	1078113 99.9%	215983 100.6%	43429 100.0%		
3F	1077949 99.9%	216147 100.6%	43488 100.2%		
4F	1079792 100.1%	216847 101.0%	43442 100.1%		
5F	1079162 100.0%	217151 101.1%	43354 99.9%		
6F	1078298 99.9%	216899 101.0%	43315.0 99.8%		
MEAN:	1078897.2 100.0%	214778.2 100.0%	43409.5 100.0%		
TPV:	916.19 0.1%	3441.97 1.6%	99.29 0.2%		
BPV:	////////// *	////////// 1.6%	////////// 0.2%	//////////	//////////

* Indistinguishable from WPV

WPV MEASUREMENTS

IN-LINE 1st	1081218 100.2%	216147 99.8%	43360 99.8%		
2nd	1078113 99.9%	216021 99.8%	43473 100.1%		
3rd	1078835 99.9%	217371 100.4%	43481 100.1%		
MEAN:	1079388.7 100.0%	216513.0 100.0%	43438.0 100.0%		
WPV:	1624.86 0.2%	745.72 0.3%	67.67 0.2%		

INLET AND EXHAUST MEASUREMENTS

Inlet	1076535	217763	43793		
Exhaust	1070797	216002	43479		
% Difference:	0.5%	0.8%	0.7%		

[Data located in appropriate daily data file]

MONITOR TYPE: GC 5890

BNW #: 32-813342

COMMENTS: Prestart tests without animals. Tests performed at original target levels before changes were made.

Data located in BNW Notebook 53435 pg 42

ENTERED BY: D. Foltz

DATE: 2/21/90

REVIEWED BY: RJ Weigel *RJW*

DATE: 3/15/90

CHAMBER UNIFORMITY DATA SHEET

COMPOUND: Chloroprene - IRTEXPOSURE ROOM NUMBER: 432

TPV MEASUREMENTS

CHAMBER: <u>125 A</u>			25 A		5A			
DATE: <u>2/17/90</u>			<u>2/17/90</u>		<u>2/17/90</u>			
SAMPLE PORT	MONITOR READING	% of Mean	MONITOR READING	% of Mean	MONITOR READING	% of Mean	MONITOR READING	% of Mean
BACK: 1B	1029047	100.1%	201840	99.9%	44053	100.3%		
2B	1036581	100.9%	201468	99.7%	44130	100.5%		
3B	1028501	100.1%	200902	99.4%	43616	99.3%		
4B	1023956	99.6%	202309	100.1%	44028	100.2%		
5B	1020590	99.3%	201836	99.9%	43991	100.1%		
6B	1027921	100.0%	201711	99.8%	44030	100.2%		
FRONT: 1F	1027285	99.9%	201373	99.6%	44167	100.5%		
2F	1027410	100.0%	203208	100.5%	44056	100.3%		
3F	1031309	100.3%	200948	99.4%	44043	100.3%		
4F	1028814	100.1%	203656	100.8%	44073	100.3%		
5F	1027540	100.0%	203187	100.5%	43468	99.0%		
6F	1024980	99.7%	203076	100.5%	43453.0	98.9%		
MEAN:	1027827.8	100.0%	202126.2	100.0%	43925.7	100.0%		
TPV:	3904.51	0.4%	943.75	0.5%	256.38	0.6%		
BPV:	//////////	*	//////////	*	//////////	0.6%	//////////	//////////

* Indistinguishable from WPV

WPV MEASUREMENTS

IN-LINE 1st	1031309	100.8%	200948	99.2%	44043	100.0%		
2nd	1013909	99.1%	203261	100.4%	44073	100.1%		
3rd	1025431	100.2%	203217	100.4%	43954	99.8%		
MEAN:	1023549.7	100.0%	202475.3	100.0%	44023.3	100.0%		
WPV:	8851.25	0.9%	1322.89	0.7%	61.89	0.1%		

INLET AND EXHAUST MEASUREMENTS

Inlet	1048547	209868	44067		
Exhaust	1019496	193611	43593		
% Difference:	2.8%	7.7%	1.1%		

[Data located in appropriate daily data file]

MONITOR TYPE: GC 5890BNW #: 32-813342COMMENTS: Prestart tests without animals. Tests performed at original target levels before changes were made.Data located in BNW Notebook 53435 pg 41ENTERED BY: D. FoltzDATE: 2/21/90REVIEWED BY: RJ Weigel *RJW*DATE: 3/15/90

APPENDIX C

DEVELOPMENTAL TOXICITY DATA

Calendar of Events
Maternal Body and Organ Weights
Reproductive Measures and Fetal Data

Calendar of Events

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits:
CALENDAR OF EVENTS

Exposure levels; treatments 1-4	0, 10, 40, 175 ppm
Ordered rabbits	11/22/89
Received rabbits (ARS#900029)	1/31/90
Assigned ID numbers to female rabbits	2/26/90
Weighed female rabbits (pre-study wt)	3/16/90
Animals released for study	3/19/90
Artificially Inseminated ♂ dg (# in Ges Grp) weighed, randomized	(A) 3/20/90 (17) (B) 3/21/90 (23) (C) 3/22/90 (23)
Exposure (6 hours/day; 7 days/week; 6-28 dg)	(A) 3/26 to 4/17/90 (B) 3/27 to 4/18/90 (C) 3/28 to 4/19/90
Weighed (6dg) started exposure	(A-C) 3/26 to 3/28/90
Weighed (15dg)	(A-C) 4/4 to 4/6/90
Weighed (22dg)	(A-C) 4/11 to 4/13/90
Weighed (29dg)	(A-C) 4/18 to 4/20/90
Teratology sacrifice (29 dg)	(A) 4/18/90 (B) 4/19/90 (C) 4/20/90
Fetal specimen exams completed	6/13/90

Maternal Body and Organ Weights

----- TMT=0 ppm Chloroprene -----

DAMID	Pre-study Wt (g)	0 dg Wt (g)	6 dg Wt (g)	15 dg Wt (g)	22 dg Wt (g)	29 dg Wt (g)	Uter Wt (g)	Liver Wt (g)	Kidney Wt (g)	Pregnant
6	4556	4555	4420	4503	4591	4727	437.30	152.55	18.38	1
9	4410	4400	4439	4620	4637	4564	613.28	116.87	21.86	1
14	4334	4336	4391	4405	4481	4616	527.90	97.11	18.22	1
15	4456	4403	4360	4329	4452	4559	9.60	145.16	24.48	0
17	4087	4103	4137	4238	4246	4406	330.42	107.18	16.47	1
29	4780	4820	4712	4742	4874	4929	6.59	158.12	20.97	0
35	4624	4544	4555	4604	4660	4832	567.44	138.78	21.64	1
36	4579	4619	4698	4853	4969	5015	551.95	136.01	24.17	1
39	4585	4533	4560	4626	4732	4720	301.30	113.20	17.71	1
44	4394	4403	4459	4402	4436	4582	497.70	119.92	24.03	1
46	4380	4403	4374	4585	4612	4624	619.53	91.10	18.73	1
52	4040	4071	4150	3960	4097	4191	9.52	139.61	19.59	0
53	4180	4184	4283	4291	4356	4471	258.10	171.40	25.85	1
54	4279	4281	4286	4416	4517	4440	474.90	97.61	19.21	1
67	3893	3944	3918	4057	4063	4282	423.94	125.86	19.88	1

Code for Pregnant Column: 1=Pregnant 0=Non-Pregnant

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Body and Organ Weights (g) for Females

----- TMT=10 ppm Chloroprene -----										
DAMID	Pre-study Wt (g)	0 dg Wt (g)	6 dg Wt (g)	15 dg Wt (g)	22 dg Wt (g)	29 dg Wt (g)	Uter Wt (g)	Liver Wt (g)	Kidney Wt (g)	Pregnant
4	4387	4430	4403	4431	4395	4448	10.16	109.47	19.93	1
5	4740	4737	4631	4698	4698	4861	174.49	186.95	22.93	1
13	4276	4301	4296	4434	4328	4057	444.90	92.64	18.18	1
19	4285	4381	4278	4267	4333	4488	9.87	114.02	15.33	0
25	4802	4870	4585	4509	4673	4695	272.61	130.41	18.53	1
26	4737	4663	4742	4851	4987	5224	717.40	137.44	19.81	1
32	4507	4559	4508	4603	4677	4799	509.75	116.65	22.00	1
33	4427	4511	4493	4401	4443	4537	10.46	121.38	16.54	1
38	4520	4433	4402	4402	4591	4837	518.28	150.63	19.58	1
55	4248	4315	4322	4353	4448	4614	543.72	119.70	19.91	1
58	4260	4275	4237	4252	4191	4170	216.60	103.41	18.00	1
59	4208	4170	4225	4317	4407	4370	567.54	80.51	16.43	1
62	4028	4077	4047	4105	4188	4188	326.19	91.55	14.46	1
63	4121	4057	4070	4224	4344	4471	464.74	120.13	18.73	1
65	4273	4381	4262	4371	4529	4557	503.09	98.55	18.06	1
68	3819	3824	3859	3884	3972	4184	202.22	130.35	17.77	1

Code for Pregnant Column: 1=Pregnant 0=Non-Pregnant

----- TMT=48 ppm Chloroprene -----

DAMID	Pre-study Wt (g)	5 dg Wt (g)	6 dg Wt (g)	15 dg Wt (g)	22 dg Wt (g)	29 dg Wt (g)	Uter Wt (g)	Liver Wt (g)	Kidney Wt (g)	Pregnant
2	4684	4557	4491	4644	4753	4915	453.78	127.88	18.98	1
7	4242	4252	4286	4417	4541	4746	498.45	141.66	19.75	1
10	4397	4377	4488	4584	4685	4594	511.58	135.77	28.32	1
12	4452	4511	4442	4582	4642	4689	415.82	116.83	21.84	1
27	4779	4713	4696	4818	4919	5185	458.72	191.19	23.71	1
31	4662	4772	4688	4749	4861	4917	351.12	148.34	22.94	1
37	4554	4519	4466	4575	4661	4738	345.21	115.86	18.64	1
41	4361	4299	4348	4455	4576	4782	584.43	138.89	28.48	1
42	4299	4255	4238	4328	4378	4485	8.98	161.44	22.91	1
43	4419	4486	4421	4456	4449	4322	282.58	116.24	28.72	1
48	4589	4549	4538	4612	4621	4854	8.75	187.35	17.37	0
50	4363	4278	4338	4446	4486	4271	158.97	189.73	17.63	1
56	3832	3836	3872	3936	3873	3987	7.51	122.79	18.81	0
64	4222	4195	4249	4351	4481	4681	435.14	181.84	22.48	1
66	4869	4856	4844	4229	4882	4171	488.88	77.87	16.37	1

Code for Pregnant Column: 1=Pregnant 0=Non-Pregnant

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Body and Organ Weights (g) for Females

----- TMT=175 ppm Chloroprene -----										
DAMID	Pre-study Wt (g)	8 dg Wt (g)	8 dg Wt (g)	15 dg Wt (g)	22 dg Wt (g)	29 dg Wt (g)	Uter Wt (g)	Liver Wt (g)	Kidney Wt (g)	Pregnant
1	4627	4631	4673	4673	4879	5107	648.61	144.38	26.26	1
3	4716	4639	4678	4805	4858	4834	586.35	95.74	17.64	1
8	4486	4398	4263	4242	4272	4361	11.36	112.17	18.38	0
18	4162	4160	4142	4263	4305	4406	533.10	93.74	18.33	1
24	4716	4668	4610	4883	4983	5021	505.21	147.02	17.96	1
28	4714	4678	4699	4810	4877	4989	395.61	138.75	23.27	1
30	4589	4542	4525	4646	4766	4976	365.27	154.96	18.42	1
34	4640	4609	4663	4793	4934	5026	504.53	135.15	22.22	1
40	4381	4368	4463	4184	4162	4218	384.25	102.89	16.97	1
45	4260	4271	4342	4459	4527	4660	556.43	108.60	17.31	1
47	4136	4190	4143	4235	4274	4355	16.37	132.94	21.18	1
49	4331	4308	4312	4475	4582	4798	585.04	127.69	23.84	1
51	4348	4329	4269	4432	4519	4426	457.03	135.07	16.56	1
60	4084	4111	4090	4180	4170	4370	399.66	117.06	17.81	1
61	4041	4059	4168	4225	4284	4495	312.41	121.36	19.14	1
69	3812	3793	3813	3936	4048	4149	471.74	80.30	17.16	1

Code for Pregnant Column: 1=Pregnant 0=Non-Pregnant

Reproductive Measures and Fetal Data

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

Code Sheet for Identification of Fetal Abnormalities

ABLL	Abnormal Lung Lobe
ACBV	Accessory Blood Vessel
FUST	Fused Sternebrae
HYIN	Hydroencephaly
LMFL	Limb Flexure
MAST	Missaligned Sternebrae
ROPB	Reduced Ossification Pelvis
ROST	Reduced Ossification Sternebrae
ROVE	Reduced Ossification Vertebrae
SHTA	Short Tail
SR0S	Supernumerary Rib Ossification Site
SRRR	Supernumerary Rudimentary Rib
SURB	Supernumerary Rib

SAS

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=8 ppm Chloroprene												
DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
6	1	1	37.542	2.8355	0.3365	1	V	SRRR				
6	2	2	.	.	.	.	.					
6	3	1	33.063	2.0535	0.2973	1	VH	ROST	MAST	SROS		
6	4	1	26.649	1.8959	0.2257	2	V	SRRR				
6	5	1	32.022	2.1550	0.2780	1	VH	ROST	SURB			
6	6	1	27.988	1.8532	0.2657	1	V	ROVE	SURB			
6	7	1	47.877	3.0909	0.4630	2	VH	ROST				
6	8	4	.	.	.	.	.					
6	9	1	37.085	2.4689	0.3522	1	V	SRRR				
6	10	1	22.315	1.3209	0.2016	1	VH	LMFL	ROST	SURB		
6	11	4	.	.	.	.	.					
6	12	2	.	.	.	.	.					
6	13	1	28.441	1.8140	0.2385	1	V	SURB				
9	1	1	47.544	3.1709	0.4077	1	V	SURB				
9	2	1	40.620	2.0606	0.3019	1	VH					
9	3	1	44.776	2.5835	0.4058	1	V	ROST	SURB			
9	4	1	45.975	2.8745	0.3511	2	VH	ROST	SURB			
9	5	1	41.546	2.0019	0.3439	2	V	ROST				
9	6	1	29.673	1.4789	0.2772	2	VH	ROST				
9	7	1	39.400	2.3133	0.3377	1	V	ROST	SRRR			
9	8	1	45.975	2.6804	0.4240	2	VH	SRRR				
9	9	1	39.814	2.2110	0.3850	2	V	ROST				
9	10	1	36.236	1.9844	0.3104	1	VH	SURB				
9	11	1	40.666	2.4692	0.4141	1	V	ROST	SROS			
14	1	1	45.255	2.7988	0.3679	1	VH	ROST				
14	2	4	.	.	.	.	.					
14	3	1	38.808	2.5293	0.3077	1	V	ROST	SRRR			
14	4	4	.	.	.	.	.					
14	5	1	40.553	2.7438	0.3412	1	VH	SURB				
14	6	1	42.433	2.8841	0.3820	1	V	ROST				
14	7	1	45.030	3.5903	0.3557	1	VH	SURB				
14	8	1	39.444	2.9019	0.3017	1	V	SURB				
14	9	1	40.809	2.8572	0.3435	1	VH					
14	10	1	40.152	3.3327	0.4289	2	V	SURB				
17	1	1	50.315	3.6719	0.3715	2	VH	SURB				
17	2	2	.	.	.	.	.					
17	3	1	44.122	2.6818	0.3126	2	V	SURB				
17	4	2	.	.	.	.	.					
17	5	1	43.557	2.8242	0.2714	1	VH	SURB				
17	6	1	45.666	3.2503	0.3593	1	V	SURB				
17	7	1	41.432	2.6276	0.3155	1	VH	SRRR				
17	8	2	.	.	.	.	.					
35	1	1	43.616	2.7527	0.4296	2	V	SURB				
35	2	1	46.084	2.9771	0.3509	2	VH	SURB				
35	3	1	53.853	4.1312	0.4591	1	V	ROST	SURB			
35	4	1	46.930	3.0957	0.4408	1	VH	SURB				
35	5	1	51.936	3.3831	0.4621	2	V					

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

----- TMT=0 ppm Chloroprene -----

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
35	6	1	53.926	3.6606	0.4975	2	VH	ROST	SURB			
35	7	1	49.921	3.5699	0.4345	1	V	SURB				
35	8	1	50.509	3.4790	0.4596	2	VH	SURB				
36	1	1	45.328	3.3355	0.3771	2	V	SURB				
36	2	1	44.870	3.1014	0.3465	1	VH	SURB				
36	3	1	42.361	2.7322	0.3654	1	V	SRRR				
36	4	1	41.778	2.7416	0.3312	1	VH	SURB				
36	5	1	43.962	3.1022	0.2778	1	V	SURB				
36	6	1	49.049	3.1928	0.3895	2	VH	SURB				
36	7	1	44.875	3.3169	0.3959	1	V	SURB				
36	8	1	45.254	3.2050	0.4148	1	VH	SURB				
36	9	1	42.761	2.9900	0.3831	1	V					
39	1	1	49.947	3.7949	0.3904	2	V	SURB				
39	2	1	52.869	3.7558	0.4532	2	VH	SURB				
39	3	1	53.257	4.1476	0.4199	2	V	ROST	SURB			
39	4	1	53.175	3.7477	0.3879	2	VH	SURB				
44	1	1	49.899	3.4874	0.4460	2	VH	SURB				
44	2	1	37.305	2.3338	0.2760	1	V	SURB				
44	3	1	33.838	1.9265	0.2738	1	VH	ROST	SURB			
44	4	1	28.319	1.6260	0.1971	2	V	SURB				
44	5	1	40.469	2.3498	0.3387	1	VH	SURB				
44	6	1	40.098	2.7018	0.3255	1	V	SURB				
44	7	1	38.489	2.2680	0.3267	1	VH	SURB				
44	8	1	40.871	3.5635	0.3990	2	V	SURB	MAST			
44	9	1	43.030	2.7325	0.3745	2	VH	SURB				
44	10	2	.	.	.	.	.					
46	1	1	42.876	3.4062	0.4235	2	V	SURB				
46	2	1	38.057	2.3940	0.3258	2	VH					
46	3	1	41.555	3.3787	0.4026	2	V	SRRR	ROST			
46	4	4	.	.	.	.	.					
46	5	1	36.608	2.2348	0.2709	2	VH	ROST	SURB			
46	6	1	34.523	1.9642	0.3441	2	V	ROST				
46	7	1	40.320	3.1549	0.4004	2	VH					
46	8	1	39.400	2.5138	0.3534	2	V	ROST				
46	9	1	35.382	2.1927	0.2925	1	VH					
46	10	1	31.977	2.0783	0.2877	1	V	ROST				
46	11	1	31.070	1.8994	0.2890	1	VH	ROST	SURB			
46	12	1	36.300	2.2530	0.3128	1	V	ROST				
53	1	1	53.120	4.2243	0.4538	2	VH	SURB				
53	2	1	51.330	4.0505	0.4020	2	V	SURB				
53	3	1	50.084	5.0334	0.5161	1	VH					
53	4	2	.	.	.	.	.					
54	1	1	39.575	2.4273	0.3527	1	VH	ACBV	ROST			
54	2	1	41.413	2.8893	0.3442	1	V	ROST	SURB			
54	3	1	39.612	2.7057	0.3518	1	VH	SURB				
54	4	1	40.704	2.5114	0.3997	1	V	ROST				
54	5	1	36.014	2.3170	0.3282	1	VH	ROST	SRRR			

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

SAS

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

----- TMT=5 ppm Chloroprene -----												
DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
54	6	1	42.582	2.9875	0.2994	1	V	ROST	SURB			
54	7	1	33.679	2.0977	0.2718	2	VH	ACBV	ROST	SURB		
54	8	1	30.521	2.0430	0.2551	2	V	ROST				
54	9	1	39.321	2.6243	0.3266	2	VH					
67	1	2	.	.	.	.	.					
67	2	1	46.216	3.3444	0.4024	1	VH	SRRR				
67	3	4	.	.	.	.	.					
67	4	1	45.014	3.4515	0.3964	2	V	ROST	SURB			
67	5	1	45.892	3.1617	0.4243	2	VH	ROST	SURB			
67	6	1	40.820	2.6716	0.3886	1	V	SURB				
67	7	1	39.877	2.7591	0.3356	2	VH					
67	8	1	35.407	2.5374	0.3214	2	V	ROST	SRRR			
67	9	1	46.471	3.4983	0.3769	1	VH	ROST	SRRR			

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
 Sex: Male = 1; Female = 2;

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=10 ppm Chloroprene

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
4	1	2										
5	1	1	56.439	4.0978	0.5286	2	VH					
5	2	1	53.584	4.1804	0.4705	1	V	SURB				
13	1	1	37.097	2.6168	0.3086	1	VH	SURB				
13	2	1	35.740	2.6217	0.2972	1	V					
13	3	1	26.894	1.6376	0.2138	2	VH	ROST	SURB			
13	4	4										
13	5	1	33.105	2.1009	0.2818	1	V	SURB				
13	6	1	41.126	3.0039	0.3347	2	VH	SRRR				
13	7	1	31.423	2.1725	0.2757	2	V					
13	8	1	32.046	2.1963	0.2973	1	VH					
13	9	1	27.418	1.8672	0.2268	1	V					
13	10	1	38.928	2.6727	0.3310	1	VH	SURB				
25	1	1	45.694	3.2442	0.3590	1	VH	SURB				
25	2	1	43.438	2.6209	0.3412	1	V	SURB				
25	3	4										
25	4	1	40.600	3.7211	0.3498	2	VH	SURB				
25	5	1	46.581	3.3455	0.4220	1	V	SURB				
26	1	1	41.831	3.2024	0.3120	2	V	ROST	SURB			
26	2	1	50.835	3.7248	0.4233	2	VH	SRRR				
26	3	1	44.957	2.8586	0.3804	2	V					
26	4	1	36.180	2.1210	0.3234	2	VH	ROST	SURB			
26	5	1	34.380	2.0774	0.2820	2	V	ROST	SRRR			
26	6	1	42.799	3.0763	0.3672	1	VH	SRRR				
26	7	1	49.795	3.9483	0.5102	1	V	ROST				
26	8	1	46.723	3.3468	0.3745	2	VH	ABLL				
26	9	1	43.902	3.1006	0.3575	2	V	ABLL	ROST			
26	10	1	40.781	2.7593	0.3495	2	VH	ABLL	SURB			
26	11	1	44.029	3.0822	0.3810	1	V	ROST	SURB			
26	12	1	37.808	2.6159	0.2963	2	VH					
32	1	1	48.172	3.2348	0.3929	2	VH	SURB				
32	2	1	44.116	2.9431	0.3656	1	V					
32	3	1	47.919	3.1151	0.3805	2	VH	ROST				
32	4	1	46.659	3.6088	0.3925	1	V	SRRR				
32	5	1	45.626	3.0607	0.3740	2	VH	SURB				
32	6	1	40.753	2.7104	0.3147	2	V	SURB				
32	7	1	49.181	3.3396	0.3947	1	VH					
32	8	1	45.784	3.1111	0.3541	2	V	SURB				
33	1	2										
38	1	1	54.547	4.5370	0.4229	2	V	SURB				
38	2	1	54.138	3.9233	0.3715	2	VH	SURB				
38	3	1	52.243	4.3604	0.3812	1	V	SURB				
38	4	1	47.441	3.4744	0.3120	1	VH	SURB				
38	5	1	53.756	4.5057	0.3669	2	V	ROST	SURB			
38	6	1	57.242	4.5805	0.4777	1	VH	SURB				
38	7	1	52.633	4.0861	0.3895	1	V	SURB				
55	1	1	47.095	3.1163	0.3843	2	VH	ROST	SRRR			

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

SAS

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=10 ppm Chloroprene

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
55	2	1	43.899	3.0975	0.3400	1	V	ROST	SURB			
55	3	1	38.810	2.2934	0.3273	1	VH	ROST	SURB			
55	4	1	42.755	2.0750	0.3823	1	V	ROST				
55	5	1	44.139	2.7577	0.4033	1	VH	ROST				
55	6	1	38.267	3.1328	0.3627	2	V	ROST	SRRR			
55	7	1	45.981	2.6435	0.3642	2	VH	ROST	SURB			
55	8	1	41.559	2.8092	0.3176	2	V	SRRR				
55	9	1	41.760	2.6292	0.3691	2	VH	ROST	SURB			
58	1	2	.	.	.	.	.	.	.	.	.	.
58	2	2	.	.	.	.	.	.	.	.	.	.
58	3	1	49.331	4.0415	0.3941	1	V	ACBV	ROST			
58	4	1	45.451	3.1928	0.3445	2	VH	SURB				
58	5	2	.	.	.	.	.	.	.	.	.	.
58	6	2	.	.	.	.	.	.	.	.	.	.
58	7	2	.	.	.	.	.	.	.	.	.	.
58	8	2	.	.	.	.	.	.	.	.	.	.
58	9	2	.	.	.	.	.	.	.	.	.	.
58	10	1	50.367	3.6008	0.2862	1	V	ROST	SURB			
59	1	1	48.805	3.3250	0.4692	1	VH	SRRR				
59	2	1	51.601	3.2694	0.4219	1	VH	SRRR				
59	3	1	52.023	3.9234	0.4349	1	V	ACBV				
59	4	1	48.794	2.8355	0.4038	1	VH					
59	5	1	43.724	3.0034	0.4245	1	V	ACBV	ROST			
59	6	1	53.868	3.7181	0.4867	2	VH	SURB				
59	7	1	40.919	2.9852	0.3637	2	V	ROST				
59	8	1	45.031	2.7722	0.3164	1	VH	ROST	SURB			
59	9	1	53.160	4.1024	0.5886	1	V	SRRR				
62	1	1	47.595	3.1297	0.4009	1	VH	SURB				
62	2	2	.	.	.	.	.	.	.	.	.	.
62	3	1	48.421	3.0287	0.4413	1	V	SURB				
62	4	2	.	.	.	.	.	.	.	.	.	.
62	5	1	50.177	3.5242	0.4540	1	VH	SURB				
62	6	1	43.175	2.8887	0.3793	1	V	SURB				
62	7	1	43.856	2.4821	0.4012	1	VH	SURB				
62	8	2	.	.	.	.	.	.	.	.	.	.
63	1	1	45.910	3.1628	0.4035	1	VH	SRRR				
63	2	1	44.050	3.0673	0.3668	2	V	ABLL	SRRR			
63	3	2	.	.	.	.	.	.	.	.	.	.
63	4	1	36.942	2.3472	0.2662	2	VH	SURB				
63	5	1	41.937	3.0540	0.3547	1	V	SURB				
63	6	1	33.239	2.1722	0.2843	1	VH	SRRR				
63	7	1	39.879	2.6984	0.3695	2	V	SURB				
63	8	1	41.519	2.7304	0.3308	2	VH	SURB				
63	9	1	43.378	3.2645	0.3197	2	V	SURB				
65	1	1	46.621	3.2661	0.4279	2	VH	ROST				
65	2	1	48.361	3.5443	0.4554	1	V	SRRR				
65	3	1	42.066	2.7885	0.3437	1	VH	SHTA	ROST	SRRR		

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

----- TMT=10 ppm Chloroprene -----												
DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
65	4	1	46.780	3.4282	0.3741	2	V					
65	5	1	33.659	1.8969	0.2915	2	VH	SURB				
65	6	2										
65	7	1	41.342	2.4941	0.3914	2	V	SURB				
65	8	1	41.911	2.7319	0.3683	2	VH	SURB				
65	9	1	41.462	2.5085	0.3556	1	V	SURB				
68	1	2										
68	2	1	48.853	3.6840	0.3994	2	VH	SURB				
68	3	2										
68	4	1	50.883	4.2166	0.4670	1	V					
68	5	2										
68	6	1	45.571	3.1193	0.3739	1	VH	ROST	SURB			

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
 Sex: Male = 1; Female = 2;

SAS

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=40 ppm Chloroprene

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
2	1	1	42.308	2.6888	0.3810	1	V					
2	2	1	44.266	2.6120	0.3911	2	VH	SURB				
2	3	1	37.829	2.1351	0.3112	2	V	ABLL				
2	4	1	38.200	2.2384	0.3225	2	VH	SRRR				
2	5	1	40.116	2.2118	0.2995	2	V					
2	6	1	37.636	2.2610	0.2888	1	VH	SHTA				
2	7	1	42.000	2.8521	0.3930	1	V	ABLL	ROVE			
2	8	1	41.750	2.3618	0.3601	1	VH	SURB				
7	1	1	42.616	3.1880	0.3483	1	VH	ROST	SURB			
7	2	1	34.098	2.0601	0.2813	1	V	SRRR				
7	3	1	40.113	2.8124	0.3185	2	VH	SURB				
7	4	1	32.652	2.1542	0.2728	2	V	SURB				
7	5	1	37.090	2.1798	0.3040	1	VH	SRRR				
7	6	2										
7	7	1	44.719	3.2327	0.4107	2	V	ROST	SURB			
7	8	1	42.828	2.5100	0.3657	1	VH	SURB				
7	9	1	45.593	3.3704	0.3282	1	V	SURB				
7	10	1	33.488	2.0956	0.2565	2	VH					
7	11	2										
10	1	1	45.967	3.0649	0.3314	1	V	SRRR				
10	2	1	46.484	2.8612	0.3371	2	VH	SRRR				
10	3	1	41.546	2.7748	0.3153	2	V	ROST	SRRR			
10	4	1	37.689	2.0651	0.2763	1	VH	SRRR				
10	5	1	30.970	1.7666	0.2299	2	V	LMFL	SURB			
10	6	1	30.525	1.8021	0.2296	2	VH	SURB				
10	7	1	39.767	2.7432	0.3511	1	V	ROST				
10	8	1	38.254	2.1631	0.3497	2	VH	ROST				
10	9	1	29.757	1.8576	0.2833	1	V					
10	10	1	32.310	1.6737	0.2602	2	VH	ROST				
12	1	1	49.195	3.6989	0.3951	2	V	SRRR				
12	2	1	44.391	2.9887	0.4094	2	VH	HYIN	ROST			
12	3	1	50.897	3.4298	0.4937	1	V	ROST				
12	4	1	50.852	3.7081	0.4620	1	VH	ROST				
12	5	1	45.575	3.2612	0.4390	2	V					
12	6	1	46.830	3.0418	0.4327	1	VH					
27	1	1	48.845	4.2001	0.3912	2	V	SURB				
27	2	1	52.876	4.1241	0.3744	2	VH	SURB				
27	3	1	41.534	3.0355	0.3396	1	V	SURB				
27	4	1	50.828	4.1507	0.4276	2	VH	ROST	SURB			
27	5	1	48.419	4.0044	0.3972	2	V	SURB				
27	6	1	27.417	2.1207	0.3302	1	VH	SURB				
27	7	1	40.945	3.1325	0.3044	1	V	SURB				
31	1	1	49.298	3.7038	0.4279	1	VH					
31	2	1	49.578	3.6353	0.4853	1	V	SRRR				
31	3	1	46.304	3.0767	0.4305	2	VH	ROST	SURB			
31	4	1	46.091	3.3020	0.4082	1	V					
31	5	1	45.938	3.2359	0.3882	1	VH	SURB				

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead

Sex: Male = 1; Female = 2;

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=40 ppm Chloroprene

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
37	1	1	49.363	3.7228	0.3969	2	VH	SURB				
37	2	1	51.427	4.3978	0.4000	2	V					
37	3	1	51.928	4.0466	0.4385	1	VH	SURB				
37	4	1	43.688	3.3638	0.3590	1	V	SURB				
37	5	1	41.454	2.8228	0.3264	2	VH	SRRR				
41	1	1	39.510	2.7106	0.3442	1	V	SURB				
41	2	1	38.418	2.2092	0.3479	2	VH	SURB				
41	3	1	48.099	3.2219	0.4115	2	V	SURB				
41	4	1	45.995	3.2746	0.4257	1	VH	SURB				
41	5	1	44.446	3.2241	0.4034	1	V	SURB				
41	6	1	43.001	2.9091	0.4404	1	VH	SURB				
41	7	1	43.533	3.1824	0.3717	2	V	SURB				
41	8	1	45.547	3.1131	0.4569	1	VH	SURB				
41	9	1	46.812	3.6427	0.3938	2	V	SURB				
41	10	1	40.479	2.6791	0.3093	2	VH	SURB				
42	1	2	.	.	.	.	.	.	.	.	.	.
43	1	1	37.416	2.7864	0.2552	1	VH	ABLL	SHTA	ROST	FUST	SURB
43	2	1	42.005	3.1472	0.3244	1	V	SURB				
43	3	1	39.448	2.7383	0.3376	1	VH	SURB				
43	4	1	40.629	2.8145	0.3156	2	V	SURB				
43	5	2	.	.	.	.	.	.	.	.	.	.
43	6	2	.	.	.	.	.	.	.	.	.	.
43	7	1	35.043	2.7007	0.3115	2	VH	ABLL	SHTA	SURB		
50	1	1	33.817	2.0912	0.2529	2	VH	SURB				
50	2	1	28.228	1.9587	0.3021	1	V	SRRR				
50	3	4	.	.	.	.	.	.	.	.	.	.
64	1	1	42.189	2.7684	0.3173	1	VH	SURB	ROPB			
64	2	1	40.258	2.6792	0.3085	2	V	ROST	SURB			
64	3	1	42.615	3.2672	0.3436	2	VH	SURB				
64	4	4	.	.	.	.	.	.	.	.	.	.
64	5	1	36.264	2.5490	0.2699	1	V	SRRR				
64	6	1	31.958	1.9722	0.2254	2	VH	SRRR				
64	7	1	53.678	4.5848	0.3013	1	V					
64	8	1	47.042	3.3159	0.3538	2	VH	SRRR				
66	1	1	49.075	3.1929	0.4486	2	VH	SRRR				
66	2	4	.	.	.	.	.	.	.	.	.	.
66	3	4	.	.	.	.	.	.	.	.	.	.
66	4	1	47.992	3.4563	0.4111	1	V	SURB				
66	5	1	42.711	2.8729	0.3783	2	VH	ROST	SRRR			
66	6	1	41.344	3.4162	0.3488	2	V	ROST	SURB			
66	7	1	42.402	2.8471	0.3782	1	VH	ROST	SURB			
66	8	1	42.105	2.9949	0.3919	2	V	SURB				
66	9	4	.	.	.	.	.	.	.	.	.	.
66	10	1	42.251	2.6428	0.3678	1	VH	ROST	SURB			

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

SAS

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=175 ppm Chloroprene

DemID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
1	1	1	47.300	3.5644	0.3548	1	VH	ROST				
1	2	1	47.023	3.8897	0.3812	1	V	ROST				
1	3	1	49.027	3.3929	0.3312	1	VH	ROST				
1	4	1	51.721	3.4036	0.3268	1	V	ABLL				
1	5	1	43.592	4.1805	0.4062	2	VH	ROST				
1	6	4	.	.	.	.	.	.				
1	7	1	39.093	2.9724	0.3549	1	V	SURB				
1	8	1	34.709	2.0006	0.2864	2	VH	ROST				
1	9	1	40.281	3.1551	0.3346	1	V	ROST	SURB			
1	10	1	51.750	4.0490	0.4952	1	VH	ROST				
3	1	1	45.791	2.8777	0.3887	2	VH	SRRR				
3	2	1	45.354	2.8241	0.3816	2	V	ROST				
3	3	1	41.605	2.6303	0.3826	2	VH	ROST	SURB			
3	4	1	43.973	2.6403	0.3526	1	V	SURB				
3	5	2	.	.	.	.	.	.				
3	6	1	29.955	1.6099	0.2267	2	VH	SURB				
3	7	1	15.519	0.5740	0.1056	1	V	ROST	SURB	ROPB		
3	8	1	25.269	1.1015	0.1835	1	VH	SURB				
3	9	1	34.641	2.2548	0.2601	2	V	SURB				
3	10	1	37.905	2.6253	0.2933	1	VH	ROST	SRRR			
3	11	2	.	.	.	.	.	.				
3	12	1	42.253	2.8596	0.3861	2	V	ROST	SURB			
3	13	1	35.444	1.9327	0.2439	1	VH	MAST	SURB			
18	1	1	49.370	3.7204	0.4489	2	VH	SURB				
18	2	1	46.140	3.4663	0.4018	1	V	SURB				
18	3	1	47.004	3.1848	0.4196	2	VH	SURB				
18	4	1	45.365	3.2441	0.4161	1	V	SURB				
18	5	1	40.750	2.7755	0.3446	2	VH	SURB				
18	6	1	39.994	2.9389	0.3697	2	V	SURB				
18	7	1	37.498	2.6286	0.3046	1	VH	SURB				
18	8	1	36.095	2.5918	0.3254	1	V	SURB				
18	9	1	47.641	3.2459	0.3790	2	VH	SURB				
24	1	1	54.831	3.7893	0.4190	2	VH	ABLL	SURB			
24	2	1	50.242	3.6545	0.3955	2	V	SRRR				
24	3	1	51.208	3.6403	0.4694	2	VH	SURB				
24	4	2	.	.	.	.	.	.				
24	5	1	51.271	4.1532	0.4207	2	V	ABLL	SURB			
24	6	1	40.030	2.6180	0.3226	1	VH					
24	7	1	49.545	3.7194	0.4219	1	V	SRRR				
24	8	1	42.647	2.6723	0.3723	1	VH					
28	1	1	46.946	3.8029	0.4217	1	V	ROST	SRRR			
28	2	1	44.000	3.3366	0.2927	2	VH	SRRR				
28	3	1	50.037	4.3439	0.4592	2	V					
28	4	1	42.280	3.2029	0.3422	2	VH	SURB				
28	5	1	44.889	3.3327	0.3896	2	V	SRRR				
28	6	1	43.738	3.2305	0.3849	2	VH	SURB				
30	1	1	55.384	3.9925	0.4298	1	VH	SURB				

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

TMT=175 ppm Chloroprene

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
30	2	1	41.674	2.4449	0.3102	2	V	SURB				
30	3	1	51.758	3.4727	0.4325	2	VH	ABLL	SRRR			
30	4	2	.	.	.	.	.					
30	5	1	55.195	4.4339	0.4470	2	V	ROST	MAST			
30	6	1	55.423	4.2070	0.4542	1	VH	ABLL	SRRR			
34	1	1	46.693	3.3217	0.3855	1	VH					
34	2	1	46.767	3.1261	0.4207	2	V	SURB				
34	3	1	44.030	3.0015	0.4452	2	VH	ROST	SRRR			
34	4	1	41.743	3.0082	0.3696	1	V	ROST	SRRR			
34	5	4	.	.	.	.	.					
34	6	1	43.871	2.9158	0.3777	2	VH	SURB				
34	7	1	43.988	2.9959	0.4427	1	V	SURB				
34	8	1	46.009	2.9598	0.4028	1	VH	SURB				
34	9	1	44.914	3.1550	0.3758	1	V	SURB				
40	1	1	49.303	3.6408	0.3541	1	VH	SRRR				
40	2	1	45.575	3.5401	0.4020	1	V	SURB				
40	3	1	35.541	2.0805	0.2978	2	VH	SRRR				
40	4	1	40.589	2.7308	0.3645	2	V					
40	5	1	40.710	2.7521	0.3366	2	VH	SURB				
40	6	2	.	.	.	.	.					
45	1	1	48.950	3.7354	0.4339	1	V	SRRR	ROST			
45	2	1	45.983	3.0429	0.3025	1	VH	ROST	SRRR			
45	3	1	44.532	3.4499	0.4584	1	V	ROST				
45	4	1	39.452	3.0010	0.3403	1	VH	SRRR				
45	5	1	42.000	2.6908	0.3897	1	V	SROS				
45	6	1	42.530	3.0060	0.3489	2	VH	SRRR				
45	7	1	43.362	3.4500	0.3430	2	V					
45	8	4	.	.	.	.	.					
45	9	1	43.676	2.9755	0.4007	2	VH	ROST				
47	1	2	.	.	.	.	.					
47	2	2	.	.	.	.	.					
47	3	2	.	.	.	.	.					
47	4	2	.	.	.	.	.					
49	1	1	46.155	3.1973	0.4123	2	V	ROST	SRRR			
49	2	1	46.000	3.0559	0.3368	2	VH	ROST	SRRR			
49	3	1	48.544	3.8814	0.4472	2	V	ROST				
49	4	1	45.867	3.3477	0.3877	2	VH	ROST	SRRR			
49	5	1	42.065	2.8144	0.2894	1	V	ROST				
49	6	1	39.824	2.8427	0.3566	1	VH	ROST				
49	7	2	.	.	.	.	.					
49	8	2	.	.	.	.	.					
49	9	1	46.177	3.5473	0.3950	2	V	ROST	SURB			
49	10	1	38.450	2.4782	0.2632	2	VH	ROST	SRRR			
49	11	1	49.110	3.4961	0.3638	1	V	SRRR	ROST			
51	1	1	41.245	2.9147	0.3663	2	VH	SURB				
51	2	1	42.580	2.9084	0.4042	1	V	ACBV	SURB			
51	3	1	46.594	3.2119	0.3761	1	VH	SURB				

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

SAS

Inhalation Developmental Toxicity Study of Chloroprene in Rabbits: Raw Rabbit Fetal Data

----- TMT=175 ppm Chloroprene -----

DamID	Site	Status	Fetal Wt(g)	Liver Wt(g)	Kidney Wt(g)	Sex	Visceral Head	Abn 1	Abn 2	Abn 3	Abn 4	Abn 5
51	4	1	40.314	2.3876	0.3233	1	V	ROST	SURB			
51	5	1	42.499	2.9555	0.3590	2	VH					
51	6	1	43.732	3.2510	0.3520	2	V	SURB				
51	7	1	30.515	1.7136	0.2880	2	VH	ROST	SURB			
51	8	1	38.935	2.3778	0.3086	2	V	SURB				
60	1	1	45.092	3.3303	0.3593	1	VH					
60	2	1	33.777	2.5124	0.2191	2	V	ROST	SURB			
60	3	1	43.859	3.1739	0.3540	2	VH	ROST				
60	4	1	36.897	2.4092	0.3100	2	V	SURB				
60	5	1	43.827	3.0293	0.3483	1	VH	SRRR				
60	6	1	33.270	2.1394	0.2735	1	V					
60	7	1	45.851	3.0503	0.4121	1	VH	SURB				
61	1	1	46.164	3.1827	0.4461	1	V	ROST				
61	2	1	34.055	2.2000	0.2447	1	VH	SRRR				
61	3	1	43.239	2.9008	0.3531	2	V	ROST				
61	4	1	47.607	3.3105	0.3899	2	VH	ACBV	SURB			
61	5	4	.	.	.	.	.					
61	6	1	48.546	3.8956	0.4800	1	V	ABLL	SURB			
69	1	1	49.493	3.2271	0.4932	2	VH	SRRR				
69	2	1	40.754	2.8810	0.2978	2	V	ROST	SURB			
69	3	1	36.188	1.8117	0.3055	1	VH	SURB				
69	4	1	42.683	3.0652	0.4173	1	V	SURB				
69	5	1	43.512	3.0481	0.3705	1	VH	SRRR				
69	6	1	40.151	3.0087	0.3752	1	V	ROST				
69	7	1	46.774	3.0110	0.4338	1	VH					
69	8	1	43.383	2.9470	0.3102	2	V	SURB				

Status: 1 = Live; 2 = Early Resorption; 4 = Late Resorption; 5 = Dead
Sex: Male = 1; Female = 2;

APPENDIX D

PROTOCOL AND CAGE MAPS

Study Protocol

Cage Maps

Study Protocol

STUDY PROTOCOL

Inhalation Developmental Toxicity Study of

Chloroprene in Rabbits

Submitted to:

Dr. Bernard Schwetz
National Toxicology Program
National Institute Environmental Health Sciences
Research Triangle Park, NC

Submitted by:

Dr. Terryl J. Mast
Battelle - Pacific Northwest Laboratory
Richland, WA 99352

March 12, 1990

I.	TITLE.....	1
II.	INTRODUCTION.....	1
III.	SPONSOR AND SPONSOR'S REPRESENTATIVE.....	3
IV.	TESTING LABORATORY.....	3
V.	PROPOSED SCHEDULE OF EVENTS.....	3
VI.	TEST SYSTEM.....	4
VII.	EXPERIMENTAL DESIGN AND DOSE LEVELS.....	4
VIII.	TEST SYSTEM HOUSING, HANDLING AND ENVIRONMENTAL CONDITIONS....	5
IX.	TEST ARTICLE.....	8
X.	DESCRIPTION OF INHALATION EXPOSURE SYSTEM.....	10
XI.	EXPERIMENTAL OBSERVATIONS.....	11
XII.	PROPOSED STATISTICAL METHODS.....	13
XIII.	RECORDS RETENTION (ØB-9A-3EØ6).....	14
XIV.	OTHER SPECIFICATIONS.....	17
XV.	HEALTH AND SAFETY.....	17
XVI.	REFERENCES.....	18
XVII.	APPROVAL BY PNL.....	19
XVIII.	APPROVAL BY NTP.....	19
XIX.	AMMENDMENTS/REVISIONS.....	19

INHALATION DEVELOPMENTAL TOXICOLOGY STUDY PROTOCOL
CHLOROPRENE

I. TITLE

INHALATION DEVELOPMENTAL TOXICOLOGY STUDY OF CHLOROPRENE IN
RABBITS.

II. INTRODUCTION

Chloroprene is a clear, colorless liquid with a pungent ethereal odor and is primarily used as a chemical intermediate in the manufacture of neoprene rubber (Table 1).

This study will determine the potential for inhaled chloroprene to cause developmental toxicity in the New Zealand white rabbit.

Table 1. Summary of Physical and Chemical Properties of Chloroprene^a

Synonym: 2-chloro-1,3-butadiene	MW: 88.54 g/mole
Molecular Formula: C ₄ H ₅ Cl	CAS No. 126-99-8
Colorless, flammable liquid. Forms explosive peroxides with oxygen.	RTECS No.: EI9625000
Polymerizes at room temperature.	Boiling Point: 59.4°C @ 760 mm Hg
Specific Gravity: 0.9583 @ 20°C	Melting Point: -130°C
Flammability Limits (by volume):	Flash Point: -20°C
Lower: 40,000 ppm	Vapor Density: 3 (air = 1)
Upper: 200,000 ppm	Permissible Exposure Limit:
Solubility:	TLV:OSHA = 25 ppm
Soluble in alcohol	ACGIH = 10 ppm
Miscible in ether, acetone, and benzene	(both with skin notations)

^aCondensed from Little (1987) and references therein.

Several studies have been conducted in order to assess the potential for β -chloroprene to cause embryonic or developmental toxicity in laboratory animals. Salnikova (1968) investigated the embryotoxic effects of inhalation exposure to chloroprene and ammonia combined; however, since a group exposed to chloroprene alone was not included it is hard to interpret their results. In a later study, Salnikova and Fomenko (1973) exposed pregnant rats to 0.016, 0.36, 0.17, 0.83, and 1.11 ppm chloroprene vapor. The length and duration of the exposures was not specified. An increased incidence of embryonic deaths was reported in the 0.83 and 1.11 ppm groups, and fetuses in the 1.11 ppm group had a significantly lower birth weight than those in the control group. Fetuses in the 1.11 ppm group were also reported to have vascular permeability disturbances as well as decreases in the lengths of the tibia and the fibula. The methodology used for the generation of the chloroprene test atmospheres and the techniques used for monitoring the purity of the atmospheres (if any) were not specified.

Culik et al. (1978) exposed CD-1 (Sprague-Dawley) rats to 0, 1, 10, or 25 ppm chloroprene, 4 h/day, on 3 through 20 days of gestation (dg) to assess developmental toxicity. Another group was exposed to the same

concentration on 1 through 12 dg in order to assess the embryotoxic potential of chloroprene. Care was taken during this study to ensure that the animals were exposed to chloroprene which was freshly distilled and relatively free of the toxic dimer. Dams exposed on 3-20 dg were killed on 21 dg and the fetuses examined for signs of developmental toxicity. Dams exposed on 1-12 dg were killed on 17 dg and the uterine contents evaluated for signs of embryotoxicity.

No clinical signs of toxicity were observed in the dams exposed to chloroprene, and there was no affect on weight gain with respect to the control animals. There was no effect on litter size, on the number of implantations, or the number of resorptions in exposed pregnant rats in either the teratology or the embryotoxicity study. Although fetal body weight was significantly greater for the controls than for the 25-ppm group, and fetal crown-rump length was significantly greater than controls for both the 10- and the 25-ppm groups, these effects were not thought to be treatment-related.

The Joint Industry Committee on Chloroprene contracted an inhalation developmental toxicity study on β -chloroprene in rats (Koeter and Appelman 1980). In this study pregnant rats were exposed to 0, 10, 25, 75, or 175 ppm chloroprene 6 hours/day, on 4-16 dg. They reported a diminished maternal weight gain during the exposure period for the 25, 75, and 175 ppm group, and during the post-exposure period for the 75 and 175 ppm groups. Post-implantation loss was significantly greater than controls in the 10, 75, and 175 ppm groups, but not in the 25 ppm group. There were fewer live fetuses in the two highest exposure groups than in the controls; however, since there was no apparent dose-response relationship the authors did not attribute this effect as related to chloroprene exposure.

There was a significant reduction in fetal weight in the 75 and 175 ppm groups, but no abnormalities attributable to treatment were found in the fetuses from any of the exposed groups. There was a statistically significant increase in the incidence of unossified forelimb phalanges and metatarsals in the 25, 75 and 175 ppm groups, and the incidence of unossified cervical vertebral bodies was increased in the 10, 75, and 175 ppm groups. The authors assert there is no clear dose-response relationship and apparently do not view these effects as being toxicologically significant since they are not reported in the study summary.

In contrast to the Russian studies (Salnikova and Fomenko 1973), chloroprene did not appear to cause developmental or reproductive toxicity at exposure concentrations as high as 25 ppm under the conditions of Culik et al. (1978). However, since a maternally toxic dose was not achieved, and 25 ppm chloroprene is the OSHA TLV, a margin of safety is not established by this study. Although the data from the Russian studies indicate that chloroprene may be considerably more toxic than implied by Culik et al. (1978), this disparity may be due to the formation of dimers and other toxic reaction products in the exposure atmospheres used by the Russians. Test material generation and monitoring methods were not described by the Russian authors although it is well known that chloroprene rapidly degrades in air to form more toxic products (von Oettingen et al. 1936, and others). In the study performed by Koeter and Appelman (1980), higher exposure concentrations were used and care was taken to ensure that the chloroprene used for exposures was freshly prepared and properly stored. Test atmospheres were not monitored for dimer concentrations in this study. Although the authors assert that the results of this study do

not indicate the presence of embryo or developmental toxicity, alternative interpretations of this data are possible.

The study in rabbits delineated in this protocol is being conducted in an attempt to clarify the ambiguities regarding the potential for β -chloroprene to cause embryonic and developmental toxicity. To date only one laboratory specie, the rat, has been well-tested for developmental toxicity. In order to facilitate an interspecies comparison of the data, exposure concentrations similar to those of Koeter and Appelman (1980) will be used in this study. The composition of the test material and the exposure atmospheres will be closely monitored for formation of the toxic dimer and permissible peroxide levels in the test material will be limited to 1 meq/kg.

III. SPONSOR AND SPONSOR'S REPRESENTATIVE

A. Sponsor:

National Institute of Environmental Health Sciences
National Toxicology Program (NTP)
P.O. Box 12233
Research Triangle Park, N.C. 27709

B. Sponsor's Representatives:

Dr. Bernard Schwetz

IV. TESTING LABORATORY

A. Facility

Battelle - Pacific Northwest Laboratory (PNL)
P.O.Box 999; Richland, Washington 99352

B. Principle Investigator:

Dr. Terryl J. Mast

V. PROPOSED SCHEDULE OF EVENTS (THIS PROPOSED SCHEDULE MAY BE ALTERED. ALL CHANGES WILL BE APPENDED TO THE PROTOCOL.)

	<u>Rabbits</u>
A. Order animals:	11/22/89
B. Animals arrive week of:	1/29/90
C. Prestart audit for GLP compliance:	3/19/90
D. Initiate breeding procedures ^a :	3/20/90
E. Initiate exposure; dg 6 ^a :	3/26/90
F. Complete exposure:	4/19/90
G. Initiate necropsy; dg 29 ^a :	4/18/90
H. Complete fetal specimen evaluation:	6/25/90
I. Submit draft report week of:	8/3/90
J. Submit final report: 45 days after receipt of reviewers comments	

^aThese dates are for the first gestational group (gestational group A), there will be three gestational groups (designated A, B, and C).

VI. TEST SYSTEM

A. Species: Rabbits

B. Strain: New Zealand white

C. Number of Animals and Supplier:

Rabbits from R & R Rabbitry, Stanwood, WA
Rabbits: 7 males
70 females

D. Age of Animals Upon Arrival:

Males: 6-7 months
Females: 5-6 months

VII. EXPERIMENTAL DESIGN AND DOSE LEVELS

A. Developmental Toxicology Study: Four groups of mated female rabbits will be exposed to the test chemical for 23 consecutive days (6-28 dg) and killed on 29 dg for maternal and fetal evaluations.

B. Exposure Regimen: Exposure concentrations of chloroprene will be 0, 10, 40, and 175 ppm, 6 hours per day, 7 days per week.

C. Experimental Animals: Approximately one-third of the females will be artificially inseminated each day during a 3-day period. Details of the artificial insemination procedure can be found in Attachment III which contains the SOP (ØB-DT-3BØE) for that procedure. On the day of insemination, females will be randomly assigned to four dose groups for a total of 16/group over 3 days (ØB-DT-3BØB). Females will be inseminated using freshly collected semen pooled from at least three breeder males. The day of insemination will be denoted as day of gestation 0 (dg 0).

E. Number of Animals in the Developmental Toxicity Study:

		Mated		Treatment			
Species		Females		Groups		Total	
Teratology	1	x	16	x	4	=	64

F. Selection of Atmospheric Concentrations: Exposure concentrations were selected on the basis of previous developmental toxicity studies in rats (Koeter and Appelman 1980), and on the lack of apparent maternal toxicity during a test exposure in pregnant New Zealand white rabbits.

VIII. TEST SYSTEM HOUSING, HANDLING AND ENVIRONMENTAL CONDITIONS

A. Quarantine (ØB-AR-3BØA)

1. The uncrating will be conducted at the door of the animal facility (LSL-II). While being removed from the crates the animals will be examined by the staff veterinarian for evidence of shipping stress. Rabbits will be transported to the quarantine room in exposure chambers.
2. The animals will be quarantined and acclimatized in the LSL-II building for 6-7 weeks prior to the start of the study. A minimum of 4 weeks is required to assure that there are no pseudo-pregnancies.
3. During the quarantine/acclimatization period the animals will be housed individually in wire cages on flush racks. The cage space will meet the requirements stated in the 1985 NIH "Guide for Care and Use of Laboratory Animals".
4. Room temperature during the quarantine period will be maintained at $69\pm4^{\circ}\text{F}$ and relative humidity at $55\pm15\%$. These measurements will be recorded at least twice daily.
5. Twelve hours light and twelve hours dark will be maintained with light starting at 0600.
6. The clinical veterinarian will make a visual inspection of the animals to be used in the study just prior to their release for the study (documented on the quarantine/acclimatization record).
7. Females not selected for the study will be discarded. The disposition of these females will be recorded on the Animal Disposition Record and retained in the study files.

B. Exposure Chamber Housing and Environmental Conditions

The exposure chamber doors will be closed throughout the exposure and non-exposure periods, except during animal care procedures.

There will be 8 rabbits per exposure chamber (2 rabbits per level) in 8 chambers. Techsorb® cageboard (Shepard Specialty Papers, Kalamazoo, MI) will be used during non-exposure periods to absorb urine.

Exposure chamber temperatures will be maintained at $69\pm4^{\circ}\text{F}$ and relative humidities at $55\pm15\%$.

Airflow will be maintained at 15 ± 3 CFM and the chamber pressure at approximately 1" water negative with respect to room pressure.

C. Feed (ØB-AR-3FØ5)

1. Purina Certified Rabbit Chow® #5322 (pellets) from A & D Brazell Distributing, Kennewick, WA, will be used during the

quarantine/acclimatization periods and throughout the duration of the study.

2. Feed will be provided, ~170 g per day, in J-type feeders during the study, except during exposure hours.

D. Water

1. Fresh softened water (ion exchange softener, Illinois Water Treatment Company, Model 2R-2240, Rockford, IL) will be supplied *ad libitum* at all times. The hardness of the water will be checked approximately once every week. Records will be retained in the LSL-II building engineer's office.
2. The automatic watering system used during quarantine will be from Systems Engineering, Napa, CA. The automatic watering system used while the rabbits are housed in the exposure chambers will be from Edstrom Industries, Waterford, WI.
3. A representative sample of animal drinking water from one of the NTP study rooms will be analyzed for contaminants at least once each calendar year (ØB-AR-3B1S).

E. Identification:

1. All animals, male and female, will be individually identified with metal eartags by the supplier prior to receipt at PNL. Prior to breeding animals will be assigned a laboratory study number. A list correlating the eartag number to the study ID number will be kept in the animal and exposure rooms at all times, and a copy will be kept with study records at all times.
2. Cage maps (ØB-DT-3BØ3) showing placement of individual animals in each cage unit of the exposure chamber will be prepared and updated as needed. Each exposure chamber will be identified by chamber number and exposure level. The proposed arrangement of the exposure chambers in the room is included in Figure 1.

F. Randomization: Females will be inseminated and assigned to exposure groups by means of a computer-assisted randomization program (ØB-DT-3BØB) which is based on a single blocking factor, 0 dg body weight.

G. Chamber, Cage, Feeder, and Automatic Water Line Sanitation Procedures

1. **Daily Sanitation Procedures (ØB-AR-3FØA)**

The excreta pans will be changed every day. The soiled pans will be hosed and washed in the exposure room.

Automatic watering systems will be checked daily during animal care procedures to ensure they are functioning properly.

2. Weekly Sanitation Procedures (ØB-AR-3BØ3, ØB-AR-3GØ1)

Chamber and cage units in use will be changed and washed every 7 days.

Exposure chambers, with animal housing components (cage units, feeders, automatic watering lines, and excreta pans), will be moved as a unit, with the chamber doors closed through the regulated (dirty) corridor, to the wash area.

The automatic water lines in the cage units and chambers will be flushed with 180-190°F water for a minimum of 1 minute prior to being washed in the cage washer.

The entire chamber will be washed in the cage washer.

The clean chamber will be stored in a clean holding area. the individual cage waterers will be checked before the chamber is moved back through the clean corridor to the exposure room.

The individual cage waterers will be checked before the chamber is used.

IX. TEST ARTICLE

A. Test Article

1. Chemical:	Chloroprene
2. Formula:	C ₄ H ₅ Cl
3. CAS No:	126-99-8
4. PNL Assigned Lot No.:	BNW53436-20 (Bottles 1-12)
5. Manufacturer:	Denka Chemical Company
8701 Park Place Blvd.
Houston, TX 77017
6. Vehicle control:	Filtered air
7. Storage conditions:	The bulk test material will be stored in the chemical transfer and storage facility immediately adjacent to the Life Sciences Laboratory-II building. The bulk test material will be maintained at approximately -20°C in amber glass bottles. A nitrogen headspace will be maintained in each bottle.

B. Bulk Assay Procedure (ØB-AC-3A14)

1. Research Triangle Institute (RTI) Assay Conclusions: Not available at this time.

PNL will confirm the identity by using infrared spectroscopy and determine the purity by using gas chromatography.

2. PNL Assay Conclusions: Infrared spectroscopy confirmed the identity of BNW# 53436-20 (bottle #2) as chloroprene. Gas chromatography showed this lot to be 98.51% pure by area percent with the major dimer constituting 0.24% of the total chloroprene sample area. Chlorobutene and cis/trans-1-chlorobutadiene constituted impurities of approximately 0.6 and 0.1%, respectively. The peroxide concentration is within the limit of 1 meq/kg.

C. Analysis Schedule

1. The identity and purity of the test material will be determined upon receipt. The purity will be performed within one month of the start of animal exposures and at 16±2 week intervals thereafter.
2. The stability of the test material in the generator will be characterized prior to the start of the study.
3. The stability of the test material in the highest and the lowest concentration exposure chambers will be characterized prior to the start of the study. This characterization will be repeated with occupied chambers following the initiation of animal exposures.
4. Peroxide concentrations in the test material will be determined within six weeks prior to the use of any particular bottle of test material. Peroxide content must be less than 1 meq/kg.

D. On-Line Gas Chromatographic (Hewlett-Packard) Chamber Monitors
(ØB-AC-3B1N, ØB-AC-3CØV)

Detector:	Flame Ionization
Column:	1' x 1/8" OD Nickel
Packing:	1% SP-1000 on 60/80 Chromosorb
Column Temperature:	120°C
Valve Oven Temperature:	140°C
Carrier:	Nitrogen
Carrier Flow:	20-30 ml/min
Sample Valve:	Hastelloy-C mounted in valve oven (Valco)
Sample Loop:	1-ml Nickel

Stream Select Valve:	12-port, Hastelloy-C mounted in valve oven (Valco).
Chromatographic Run Time:	Approximately 1.5 min exclusive of report generation.
On-Line Standard:	Chloroprene in nitrogen will be used to detect drift of the online monitor.
Calibration Frequency:	Daily check against on-line standard, additional checks against grab samples as indicated by the on-line standard.
Control Range:	Target concentration $\pm 10\%$.
Critical Limits:	Target concentration $\pm 20\%$, alarmed (will be automatically shut off if upper limit is reached).
Monitoring Frequency:	Each chamber and the room will be monitored approximately once every 30 minutes. The concentration of the major chloroprene dimer in the distribution line will be monitored daily.

X. DESCRIPTION OF INHALATION EXPOSURE SYSTEM

The inhalation chambers will be located in room 432 of the LSL-II building. A detailed description of the inhalation exposure system to be used in this study is included in Attachment 2 of this protocol. The location of the exposure room and chamber layout are shown in Figure 1.

A. Environmental Monitoring

1. Air filtration: HEPA and charcoal filters will be used for intake air, and a HEPA filter will be used for exhaust air. New exhaust and intake filters will be installed prior to the start of the study.
2. Temperatures will be monitored by resistance temperature devices (RTDs) multiplexed to a digital thermometer with computer data acquisition at approximately 4-hour cycles for 24 hours per day (minimum of 3 measurements per day). The control range is $69 \pm 4^\circ \text{F}$ with critical limits, < 63 or $> 75^\circ \text{F}$. Any chamber temperature excursion beyond the critical limits will be recorded and alarmed automatically.

3. Relative humidity will be monitored by a single dew point hygrometer in conjunction with a multiplexed sampling system with computer data acquisition at approximately 4-hour cycles, 24 hours per day (minimum of 3 measurements per day). The control range is $55 \pm 15\%$ with critical limits of $<35\%$ or $>75\%$. Any relative humidity excursion beyond the critical limits will be recorded and alarmed automatically.
 4. Chamber airflow will be monitored at an exhaust orifice using a multiplexed Validyne pressure transducer system with computer data acquisition at approximately 4-hour cycles, 24 hours per day (minimum of 3 measurements per day). The control range is 15 ± 3 air changes/hour (15 ± 3 CFM) with critical limits of <10 CFM or >20 CFM. Any chamber flow excursion beyond critical limits will be recorded and alarmed automatically. A critically low flow will result in automatic termination of the exposure. Chloroprene concentrations in the chambers may be controlled in part by the adjustment of chamber airflow.
 5. Chamber vacuum will be monitored using a multiplexed Validyne pressure transducer system with computer data acquisition at approximately 4-hour cycles, 24 hours per day (minimum of one measurement per day). The control range is -0.2 to -2.0 inches of water pressure with critical limits set at the same values. Any chamber vacuum excursion beyond the critical limits will be recorded and alarmed automatically. If chamber vacuum exceeds the limits of -0.2 inch of water, the exposure will be automatically terminated.
 6. Uniformity of the concentration of the test chemical in each of the chambers will be made at the beginning of the study with the animals in the chambers. A between port and within port variability of $\leq 5\%$ relative standard deviation (RSD) is considered acceptable.
 7. Build up and decay times will be determined during the pre-start testing period. Buildup and decay measurements will also be made at the beginning of this study with the animals in the chamber.
 8. Samples will be taken from all chambers to check for the presence of aerosols before the start of exposures and at the beginning of the study when animals are present.
- B. Effluent Treatment (ØB-AC-3A37) Chamber exhaust will be HEPA filtered to remove all aerosol. The building exhaust stack will be monitored once during the study to prove efficiency of the effluent treatment.

XI. EXPERIMENTAL OBSERVATIONS

- A. Clinical Observations (ØB-DT-3BØ3): Study females will be observed twice daily for mortality, morbidity, and signs of toxicity. The date and time of death or euthanasia of moribund animals will be recorded and the animals will be necropsied (ØB-DT-3BØF).

- B. Body Weights (ØB-AR-3GØH): All females will be weighed within one week prior to mating. Mated females will be weighed on 0, 6, 15, 22 and 29 dg.
- C. Scheduled Necropsy (ØB-DT-3BØG): Animals will be weighed in the exposure room the morning of sacrifice. They will then be transported to room 1428 and killed by a lethal injection of sodium pentobarbital (Nembutal®).

At necropsy maternal animals will be examined for gross tissue abnormalities. In order to document the presence of lesions which may be due to chemical exposure, any organs or tissues with lesions will be preserved in 10% neutral buffered formalin (NBF). In this case, comparable organs or tissues from approximately 20% of the control animals will be preserved in 10% NBF. Liver and kidneys will be weighed and tissues discarded unless abnormal.

The gravid uteri will be removed immediately upon death of the animals. The gravid uteri will be weighed, opened, and the number, position and status of implants will be recorded. Any apparently non-gravid uteri will be weighed and stained with 10% ammonium sulfide to detect possible implantation sites. The placentas will be examined and preserved in 10% neutral buffered formalin (NBF) if abnormal. Ovarian corpora lutea counts will be obtained for both gravid and non-gravid females.

D. Fetal Examination (ØB-DT-3BØG):

1. The identity of live fetuses (by study, dam number, and implant number) will be retained throughout all examinations. Live fetuses will be examined for gross defects and weighed (ØS-SI-5EØ2, ØS-SI-5EØ3). Following euthanasia, a complete visceral examination will be performed on 100% of all live fetuses. Sex will be determined on all live fetuses by internal examination of gonads. Liver and kidneys will be weighed and tissues discarded unless abnormal.
2. Approximately 50% of the fetal heads will be removed and examined by razor-blade sectioning of the fixed preparations (ØB-DT-3BØI). All carcasses, with and without heads, will be stained with Alizarin red S and alcian blue and examined for cartilage and centers of ossification (ØB-DT-3BØH). Records of morphologic lesions observed during examinations will include photographs of unusual representative lesions. All fetal specimens will be double bagged and sent to NTP Archives. (ØB-HI-3GØ9, ØB-DT-3FØ2)

F. Indices of Effects: The following parameters will be expressed as mean $\pm$ SD, when appropriate.

- Number of animals on study, number of dead animals, animals removed from the study and reason for removal
- Summary of observed toxicity, including incidence of changes detected during clinical observations
- Number and percent pregnant at sacrifice
- Maternal body weights taken on 0, 6, 15, 22 and 29 dg (sacrifice)
- Weight of gravid uterus
- Maternal liver and kidney weights
- Extragestational weight and weight gain
- Number of corpora lutea/dam
- Number of implantation sites/litter
- Number of litters with live fetuses
- Number and percent of live fetuses/litter
- Body weight of male and female fetuses/litter
- Fetal liver and kidney weights
- Sex ratio of fetuses/litter
- Number and percent of early and late resorptions/litter
- Number and percent of dead fetuses/litter
- Number and percent of non-live/litter (early and late resorptions and dead fetuses)
- Listing of malformations and variations observed in fetuses/litters
- Number and percent fetuses/litter with malformations (variations)
- Number and percent of litters with malformations (variations)

XII. PROPOSED STATISTICAL METHODS

The methods proposed for the statistical analyses of representative maternal, reproductive and fetal indices of effects are: summary statistics, N, mean, standard deviation, with accompanying ANOVA based on multiple comparisons where appropriate. Arc sine transformations will be performed on proportional incidence data. Further statistical analyses will be performed as necessary.

XIII. RECORDS RETENTION (OB-9A-3E06)

Records that accumulate during the study will be retained at PNL until requested and shipped to NTP archives. Some of these records may be presented as part of the protocol or reports. These will include but not be limited to the following records:

A. Personnel Records

1. List of PNL personnel participating in the study
2. Name, address, and function of any outside consultant(s)
3. Record of removal of any individual from direct contact of the test system due to illness

B. Health and Safety Records (original records and five copies of microfiche will be submitted to NTP within approximately two months after the end of each fiscal year). Chemical specific records will be submitted with the study. Facility specific records will be submitted annually.

1. Medical records of all personnel participating in the study. These records will be retained by Hanford Environmental Health Foundation (HEHF), P.O. Box 100, Richland, WA 99352 for a minimum of 40 years. A letter verifying this arrangement will be retained for each test material file.
2. Records and results of any biological monitoring on laboratory personnel (if applicable)
3. NTP Health and Safety package for chloroprene
4. PNL biohazard protocols and PNL Health and Safety Plan
5. Chemical specific health and safety training records
6. Waste disposal records
7. Respiratory protection program with documentation of user training (specific fit testing if needed) for each type of respirator
8. Building ventilation system, hoods and exhausting system monitoring records (pertinent to NTP studies)
9. Health and safety section of the monthly progress reports
10. Accident/injury reports for personnel involved in this study
11. NTP site visit reports, attention items and related correspondence on health and safety

C. Protocols

1. Approved and dated PNL study protocol

AIM

Association for Information and Image Management

1100 Wayne Avenue, Suite 1100
Silver Spring, Maryland 20910

301/587-8202

Centimeter

Inches

MANUFACTURED TO AIM STANDARDS
BY APPLIED IMAGE, INC.

3 of 3

2. Protocol amendments including NTP technical contract modifications which affect the study
3. Documentation of any deviation from the protocol
4. Documentation of any unforeseen circumstances that may affect the integrity of the study and corrective actions taken.

D. Test Material Records

1. Test material identity records including manufacturer, quantity, lot number(s), purity grade and date(s), etc.
2. NTP analytical contractor characterization reports
3. NTP analytical contractor bulk stability reports
4. NTP analytical contractor shipment records (if available)
5. PNL test chemical receipt records
6. PNL storage records including storage conditions
7. PNL bulk analysis and degradation records
8. PNL method development records
9. Chemical exposure generation system description and procedures
10. Chamber concentration monitoring records
11. Uniformity (chamber balance) records
12. Generation and chamber degradation study records
13. PNL test material inventory and usage records
14. Records of shipment to NTP repository of any unused test material
15. Aerosol determination records
16. Chamber concentration buildup, decay, and overnight monitoring records
17. Exposure generation operating parameter records

E. Animal Records - Pretest

1. Animal receiving records including supplier, species, strain, birth week, sex, number of animals for each sex, receiving date and receiving conditions.
2. Quarantine records
3. Pretest health records and animal health notebook
4. Randomization records

5. Animal identification records
6. Written release records from clinical veterinarian
7. Disposal of excess animals

F. Animal Records - On Test

1. Exposure room location and chamber layout records
2. Chamber cage map
3. Cage type, rack type during study
4. Cageboard type
5. Type of watering system
6. Body weight records
7. Daily observation records
8. Clinical signs of toxicity records
9. Study animal disposition records
10. Inventory list of archived specimens

G. Feed

1. Feed tags with manufacturer, lot numbers and milling dates
2. Feed analysis records as provided by NTP analytical contract laboratory

H. Water

1. Annual water analysis
2. Weekly water hardness check (records will be maintained in building engineer and/or building manager's office).

I. Quarantine Room, Exposure Room, and Inhalation Exposure Chamber Records

1. Exposure chamber description
2. Exposure suite control center description
3. Temperature raw data and daily and monthly summation report
4. Relative humidity raw data and daily and monthly summation reports
5. Airflow raw data and daily and monthly summation reports
6. Chamber vacuum raw data and daily and monthly summation reports

7. Exposure system monitors calibration and maintenance records.
8. Description of the lighting system and light/dark regimen.
9. Sanitation procedures and pest control program.

J. All Relevant Correspondence

K. Reports

1. Monthly Progress Report
2. Special study reports (if any)
3. Incident reports (if applicable)
4. Draft study report
5. Final study report

L. Internal Computer Generated Forms and Tables

1. Developmental toxicology results and statistical analyses
2. Analytical chemistry results
3. Exposure suite control center computer printouts
4. XYBION printouts (if any)

XIV. OTHER SPECIFICATIONS

- A. This study will be performed in compliance with the FDA Good Laboratory Practice Regulations for nonclinical laboratory studies (21 CFR 58) except where deviations are required by the NTP April, 1987 General Statement of Work and subsequent modifications.
- B. This protocol will be the controlling document in case of discrepancies between the protocol and SOPs. If this occurs the Principal Investigator is to be notified immediately for clarification.
- C. A list of all relevant standard operating procedures (SOPs) for this study are present in Attachment I.

XV. HEALTH AND SAFETY

PNL's Health and Safety Plan (ØB-HS-3S1C) has been approved by NTP. In addition, a respiratory program is instituted. This is supplemented by using supplied-air respirators (ØB-HS-3S19) which will be worn by personnel at all times when they are in exposure rooms and by having available self-contained breathing apparatus (ØB-HS-3SØW) for use when entering a room under emergency conditions following a leak.

XVI. REFERENCES

- Culik R., D.P. Kelly, J.J. Clary. 1978. Inhalation studies to evaluate the teratogenic and embryotoxic potential of β -chloroprene (2-chloro-butadiene-1,3). Toxicol. Appl. Pharmacol. 44:81-88.
- Koeter, H.B.W.M., L.M. Appelman. 1980. Inhalation embryotoxicity/teratogenicity studies with β -chloroprene in rats. Report No. R6387. Centraal Instituut voor Voedingstonderzoek, Utrechtseweg 48, Zeist.
- Salnikova, L.S. 1968. Embryotropic effect of volatile substances from nairit latexes. Toksikol. Nov. Prom. Khim. Veshchestv. 11:106-111.
- Salnikova, L.S., V.N. Fomenko. 1973. Effect of chloroprene on embryogenesis. Gig. Tr. Prof. Zabol. 8:23-26.
- von Oettingen, W.F., W.C. Hueper, G. Deichmann, F.M. Wiley. 1936. 2-Chloro-butadiene (chloroprene): Its toxicity and pathology and the mechanism of its action. J. Ind. Hyg. Tox. 18:240-270.

XVII. APPROVAL BY PNL

Terryl J. Mast

Terryl J. Mast, PhD
Principle Investigator

3/13/90

Date

Patricia S. Rueninler
Quality Assurance Officer

3/13/90

Date

XVIII. APPROVAL BY NTP

Bernard A. Schwetz

Bernard A. Schwetz, DVM, PhD
Study Officer

20 Mar 90

Date

XIX. AMMENDMENTS/REVISIONS

STANDARD OPERATING PROCEDURES
FOR DEVELOPMENTAL TOXICOLOGY STUDIES

DEVELOPMENTAL TOXICOLOGY

ØB-DT-3BØ3	Cage Location Maps and Daily Observations
ØB-DT-3BØB	Randomization of Animals
ØB-DT-3BØE	Artificial Insemination of Rabbits
ØB-DT-3BØF	Necropsies of Dead or Moribund Animals
ØB-DT-3BØG	Developmental Evaluations for Teratology Studies
ØB-DT-3BØI	Examination of Fetal Heads Fixed in Bouin's Solution
ØB-DT-3BØH	Skeletal Examination of Fetuses Using Alizarin Red
ØB-DT-3FØ2	Shipping Developmental Toxicology Materials
ØB-HI-3GØ9	Operation of Sealer
ØS-SI-5EØ2	Macintosh Weighing Programs
ØS-SI-3EØ3	Data Transfer from Macintosh to VAX Using MacTerminal

ANIMAL FACILITIES

ØB-AR-3BØ3	Handling and Changing Out Exposure Chambers and Cage Units
ØB-AR-3BØD	Handling, Changing, and Storage of Animal Cages and Racks
ØB-AR-3BØG	Barrier Procedures for LSL-II Animal Facility
ØB-AR-3B1S	Monitoring for Bacterial Contamination in Animal Drinking Water
ØB-AR-3BØA	Quarantine of Rabbits
ØB-AR-3FØ5	Management of Animal Feed
ØB-AR-3FØA	Daily Care of Bioassay Animals and Cleaning of Exposure Rooms
ØB-AR-3GØ1	Pre-Cleaning Equipment and Operation of Cage-Bottle and Rack Washers
ØB-AR-3GØH	Rodent Weighing using Toledo 8142 Automatic System
ØB-AR-3HØ1	Bi-Weekly Deep Cleaning of Exposure Rooms and Occupied Animal Rooms

INHALATION EXPOSURE AND BIOENGINEERING

ØB-BE-3B1X	Relative Humidity Determination Via Use of Dewpoint Hygrometer
ØB-BE-3B24	Inhalation Exposure Chamber Balance
ØB-BE-3B3H	Build-up and Decay and Overnight Concentration Monitoring
ØB-BE-3B2V	Chloroprene Exposure System Daily Operating Procedure
ØB-BE-3CØJ	EG&G Hygrometer: Operation, Maintenance, and Calibration
ØB-BE-3CØL	RTD Thermometer Calibration
ØB-BE-3CØV	Calibration and Check of Chamber Airflow Using Digital Anemometer
ØB-BE-3DØ6	Chamber Leak Test
ØB-BE-3DØE	Exposure Suite QC, Maintenance, and Calibration
ØB-BE-3DØK	Chloroprene Exposure System QC, Maintainance and Calibration
ØB-BE-5EØ1	Exposure Suite Computer Program Description
ØB-BE-3EØB	Exposure Suite Data Analysis Program Operation
ØB-BE-3EØE	Exposure Suite Data Editing Program Operation
ØB-BE-3GØ4	Exposure Suite Routine Computer Operation

ANALYTICAL CHEMISTRY

ØB-AC-3A14	Bulk Analysis of Chloroprene
ØB-AC-3A37	Analysis of Building Exhaust for Chloroprene
ØB-AC-3CØV	Chloroprene - Calibration of Chamber Monitor
ØB-AC-3A18	Analysis of Bulk Chloroprene for Peroxide
ØB-AC-3A19	Analysis of Chloroprene for Dimer
ØB-AC-3B1N	Chloroprene - Operation of HP5890 GC for Monitoring
ØB-AC-3BIT	Chloroprene Chemical Handling
ØB-AC-3DØ9	Maintenance of the Hewlett Packard Model 5890 Gas Chromatograph

SAFETY

ØB-HS-3S19 The 3M Brand W-2860 Hardcap, Continuous-Flow Air Line Respirator

ØB-HS-3S1B Bioassay Studies: Respiratory Protection Program

ØB-HS-3S1C Bioassay Studies: Health and Safety Plan

ØB-HS-3S1L Biohazard Protocol - Chloroprene

ØB-HS-3SØW Scott 4.5 Self-Contained Breathing Apparatus (SCBA)

NTP PROJECT OFFICE

ØB-QA-3EØA Filling Out Data Sheets

ØB-9A-3EØ6 Data Handling and Storage of NTP Study Documents and Materials

INHALATION EXPOSURE SYSTEM DESCRIPTION

I.	ANIMAL EXPOSURE CHAMBER_____	II-2
II.	EXPOSURE SUITE SYSTEM DESCRIPTION _____	II-2
III.	TEST ARTICLE GENERATION AND MONITORING	
A.	Test Article Generation System _____	II-3
B.	Test Article Monitoring System _____	II-4
IV.	CHAMBER ENVIRONMENTAL CONTROL AND MONITORING	
A.	Temperature _____	II-5
B.	Relative Humidity _____	II-6
C.	Air Flow _____	II-7
D.	Vacuum _____	II-7
V.	ANIMAL FACILITY AIR HANDLING SYSTEM _____	II-8
VI.	EXPOSURE SUITE ALARM SYSTEM _____	II-8
VII.	CHAMBER EXHAUST WASTE TREATMENT _____	II-9
VIII.	DATA HANDLING _____	II-9
IX.	EQUIPMENT OR POWER FAILURE PROTECTION SYSTEMS _____	II-10
	REFERENCES _____	II-11
	FIGURES _____	II-12

INHALATION EXPOSURE SYSTEM DESCRIPTION

I. ANIMAL EXPOSURE CHAMBER

The animals will be exposed and maintained in inhalation exposure chambers developed at BNW (U.S. Patent No. 4,261,741, August 12, 1980; Moss, 1980; Brown and Moss, 1981; Moss *et al.* 1982) and now commercially produced by the Harford Division of Lab Products, Inc., Aberdeen, MD. The chamber (Figure 1) facilitates multiple-tier exposures of various laboratory animal species to aerosol- and vapor-laden atmospheres. The total volume of the chamber is 2.3 m³ with an active mixing volume of 1.7 m³, the remainder being the inlet and exhaust volumes where animals are not placed. For rabbits, there are two levels of caging, each level split into two tiers which are offset from each other and from the chamber walls. Drawer-like stainless steel cage units composed of two animal cages are suspended in the space above each tier. Stainless steel catch pans for the collection of urine and feces are suspended below each cage unit. Catch pans are left in position during each exposure period. Cageboards are added to these catch pans during non-exposure periods to reduce ammonia concentrations.

The chamber was designed so that uniform aerosol or vapor concentrations can be maintained throughout the chamber when the catch pans are left in position. Incoming air containing a uniform mixture of test material is diverted so that it flows vertically along the inner surfaces of the chamber. Eddies are formed (Figure 1) at each tier as the aerosol or vapor flows past the catch pans. Stagnant zones that would normally exist above each pair of catch pans are cleared by exhaust flow through the space between the tiers. Aerosol or vapor reaching the lowest level is deflected across the bottom tiers by metal strips in the space between the catch pan and the wall. Tests have shown that aerosol or vapor concentration homogeneous to within 8% throughout the chamber can be obtained repeatedly provided the aerosol or vapor is uniformly mixed before passing through the chamber inlet (Griffis *et al.* 1981).

II. EXPOSURE SUITE SYSTEM DESCRIPTION

The exposures will be conducted using an automated data acquisition and control system in an exposure suite (Figure 2) consisting of three exposure rooms and a Suite Control Center room (only one of the exposure rooms will be used for this study). A central computer monitors the basic chamber functions (i.e., test chemical concentration, air flow, vacuum, temperature, and relative humidity) in each of the three exposure rooms. The executive computer is a Hewlett-Packard (HP) Model 9816. All data acquisition and system control originates from this computer.

All experimental protocols related to the data acquisition and control system (such as data channel assignments, monitoring frequencies and alarm settings) reside in the executive computer and are entered into tables accessed by menus.

Data input to the executive computer is accomplished through several interface instruments. All on-line chemical monitor data are collected and preconditioned by HP-85B computers, one for each of the exposure rooms. Conditioned data are transferred to the

executive computer for analysis, concentration control, data storage, and printing. Data from all monitoring equipment other than chemical monitors are inputted through a Colorado Data Systems (CDS) Model 53A-IBX Intelligent Interface System (IIS).

System control is provided from the computer by means of control relays in the CDS IIS. These relays control such devices as valves, drive motors, audible alarms, indicator lamps, etc.

Data and comments from each exposure room are stored on separate magnetic diskettes by HP Model 9121 micro-floppy disk drives. Data and comments from each exposure room are printed on separate thermal dot matrix printers (HP Model 2671G). Data are printed and stored immediately upon completion of the measurement to the "Daily Log". At the end of the day (24-hour period), the daily data are analyzed and a summary is printed. This summary includes the mean, standard deviation, % relative standard deviation, maximum, minimum, and number of measurements for each set of data for the 24-hour period. A second printout provides a table of outliers (i.e., all data points which are beyond the critical limits defined in the protocol). This outlier table allows rapid determination of problem areas in the data. A third printout provides a list of all comments generated by the computer and operators. This allows rapid inspection of generation start and stop times and any problem areas.

A complete description of the software for this system is contained in BNW document ØB-BE-5EØ1. Maintenance of this system is detailed in SOP # ØB-BE-3DØE. Routine operation of the system software and hardware are detailed in SOP # ØB-BE-3GØ4.

III. TEST ARTICLE GENERATION AND MONITORING

A. Test Article Generation System

The method used for the generation and delivery of chloroprene (CHLP) is shown in Figure 3. The generator is housed in a vented cabinet located in the Suite Control Center. The cabinet is vented to the building exhaust. The generator employs a rotary evaporation system (Büchi Rotavapor, Model EL-131S) to produce high purity chloroprene vapor. A 1-liter evaporating flask is filled (under a nitrogen blanket) once at the beginning of each day with chloroprene from one or more of the original chloroprene containers. The flask is immersed in a hot water bath at approximately 150°F. A calibrated nitrogen stream is metered into the rotating flask and through the generator from the flask to the condenser. The resulting vapor is forced out of the mouth of the flask into the condenser. The condenser temperature is maintained by a temperature-regulated water bath. The condenser brings the vapor to the desired temperature and condenses part of the total vapor. The condensation controls the chloroprene vapor concentration produced by the generator. Condensate is returned to the rotating flask.

The nitrogen flow and temperature of the circulating water and other parts of the vaporization system can be adjusted until a sufficient output of vapor is reached. The total chloroprene output of the generator can be calculated from the metered nitrogen flow and an estimate of the chloroprene vapor pressure at the minimum temperature reached within the condenser. For example, at a circulating water temperature of 28° F, the concentration of the vapor is estimated at 89,000 ppm. The generator output is set to meet the demands of achieving the target concentrations in the exposure chambers plus a 20% surplus delivery line flow. The excess delivery flow allows for adjustments to the vapor delivery system's operating parameters without affecting other flow rates. Unused delivery line flow is diluted with compressed air and then sampled by a second GC before being vented to waste.

From the condensing column, the vapor enters a short distribution manifold from which individual delivery lines carry a metered amount of vapor to each exposure chamber. Each delivery line is connected to the distribution manifold through a fine metering valve and flowmeter. At the chamber end of each delivery line, an AirVac® pump withdraws the appropriate amount of chemical vapor from the manifold to reach the target concentration. Chamber concentrations are adjusted by the metering valves and/or by adjustment of the compressed air pressure to the vacuum pumps. At the end of the delivery line, the vapor enters a pneumatic 3-way valve where it is directed to the exposure chamber or the chamber exhaust system as appropriate. These valves allow for a faster buildup of vapor concentration at start-up as well as more rapid diversion of chemical flow from chambers in the event rapid shutdown is necessary. At the start of a generation period, these valves are positioned to route the vapors to the chamber exhaust until the concentrations in the distribution system have stabilized and volatile trace contaminants in the evaporation flask are purged. When the system is stable and exposures are ready to proceed, computer-controlled start-up of the exposure is initiated by rotation of the 3-way valves to the exposure position. Dilution of the nitrogen/chloroprene vapor mix with air to the target chamber concentrations occurs immediately before entry to the chambers.

Generation proceeds in stages: warm-up, generation and shutdown. During warm-up, the pneumatic 3-way valves are in the exhaust position and heating of chemical in the evaporating flask and water in the bath proceed together. After operating temperatures are reached, at the direction of the operator, computer-controlled start-up is initiated and the pneumatic valves are set to the exposure position. After stabilization of the chamber concentrations and possible fine adjustments of the vapor delivery and nitrogen flow rates, the generator is in its normal operating stage. During this stage, the generator operates under computer and exposure-operator supervision. The exposure operator monitors the various vaporization temperatures as well as other operating parameters. The computer can initiate an interruption of chemical delivery to any chamber under certain conditions by rotating the appropriate 3-way valve.

Shutdown at the end of the generation stage is similar to a computer-initiated shutdown. After the 3-way valves are rotated to the exhaust position, stopping exposures, the operator stops the evaporator flask rotation, turns off the bath heater and floods the bath with cold water. After the evaporating flask cools, it is removed from the generator and replaced with a cleaned flask. Nitrogen continuously purges the vaporization system until preparation for the next day's generation.

Routine operation of the generation system is detailed in SOP# ØB-BE-3B2V. Maintenance and QC procedures are detailed in SOP# ØB-BE-3DØK.

Measurement of vapor concentration versus time (buildup and decays) will be obtained for all chambers before the start of the study and during the study. (SOP# ØB-BE-3B3H)

B. Test Article Concentration Monitoring

An on-line Hewlett-Packard Model 5890 gas chromatograph equipped with an flame detector (FID) will be used to monitor the exposure chambers, the control chambers, the exposure room, and the output of a standard gas cylinder. Sampling from multiple positions will be accomplished by means of an oven-mounted, 12-port stream select valve. The sampling system (Figure 4) is incorporated into the relative humidity (RH) sampling system. Samples of the atmosphere from each sample location are continuously drawn by a vacuum pump through teflon sample lines to a location near the input to the 12-port sample valve. This assures fresh samples at the monitor. The sample lines, which continue from the point where they tee-off to the 12-port valve to the dewpoint monitor, are 1/16" teflon.

Sample values are accumulated and printed by an HP Model 85B computer until samples from all twelve ports of the stream select valve have been measured. These values are then sent to the executive computer for printing and storage. Each value accumulated by the HP85B is compared with limit values for that particular location. If the value is beyond the control limits, the HP85B will immediately send the information to the executive computer which will take the appropriate action as follows:

- Concentration $\geq$ non-critical low limit and $\leq$ non-critical high limit:
No action
- Concentration $<$ non-critical low limit but $\geq$ critical low limit:
Indicate on daily printout
- Concentration $<$ critical low limit:
Indicate on daily printout and activate audible alarm
- Concentration $>$ non-critical high limit but $\leq$ critical high limit:
Indicate on daily printout
- Concentration $>$ critical high limit:
Indicate on daily printout, activate audible alarm, terminate generation of test material

The on-line gas chromatograph will be calibrated against quantitatively analyzed bubbler grab samples collected from the exposure chambers. Standards will be prepared gravimetrically. The on-line monitor calibration will be confirmed at least once per month by grab sampling. The calibration check will be performed according to SOP #ØB-AC-3CØV. Instrument drift will be checked daily against an on-line standard.

A constant concentration stream of chloroprene vapor, used only to detect instrument drift, is provided by compressed nitrogen/CHLP bottled gas. The chloroprene standard is monitored every 12th sample throughout the exposure period. The standard is also checked before the start of each exposure day. The measured concentration for the standard is required to be within $\pm 10\%$ of the assigned target value before any exposure begins without consultation with the exposure control task leader. During the course of the exposure, if the on-line standard is within $\pm 5\%$ of the target value, no change in calibration is required. If the on-line standard is beyond $\pm 5\%$ of its assigned target, the lead chemist is informed immediately by an exposure specialist. Additional grab sampling is initiated by the chemist on a case by case basis when drift in the on-line standard is between $\pm 5-10\%$. A cumulative drift of 10% or greater requires immediate recalibration of the on-line monitor by grab sampling.

Daily operating procedures for the concentration monitoring system are contained in SOP #ØB-AC-3B1N. Routine maintenance of the GC is covered in SOP #ØB-AC-3DØ9.

The uniformity of the distribution of the test chemical in the chamber will be checked before the start of the study and during the first week of exposure. (SOP #ØB-BE-3B24)

IV. CHAMBER ENVIRONMENTAL CONTROL AND MONITORING

A. Temperature

Nearly all of the heat load contributed to the exposure chamber by the animals is dissipated from the chamber by radiation through the chamber wall (Bernstein and Drew, 1980). The temperature of the air supplied to the chamber has little effect on the temperature of the chamber. On the other hand, the temperature of the room housing the chamber has a great deal

of effect. Consequently, chamber temperature is maintained in the proper range primarily by controlling the room temperature.

Temperatures of the exposure chambers and exposure room are measured by Resistance Temperature Devices (RTDs). The RTDs are placed in a representative location in each chamber. Each RTD is connected to an Omega Model 412B Digital Thermometer by either a manual select switch or by computer controlled scanner relays. This allows the temperature to be read manually or to be recorded automatically by the exposure system executive computer. Temperatures are automatically recorded at regular intervals during each 24-hour day. All temperature measurement equipment except the RTDs is located in the Exposure Suite Control Center. RTDs are calibrated to within 0.5°F of a certified mercury thermometer (SOP # ØB-BE-3CØL) before the start of the study and at least every two months thereafter.

B. Relative Humidity

Relative humidity (RH) in the exposure chambers is controlled by the system depicted in Figure 5. Equipment located in the RH Control Equipment Room (Room 335 of the LSL-II basement) provides separate ducts of dry and moist air to each exposure chamber. Filtered air with a maximum dewpoint of about 53°F is supplied to the RH Control Equipment by the building HVAC system. This air is evenly delivered to two ducts. Air from the first duct passes into a plenum where steam, generated from city tap water with no additional additives, is injected to bring the air to a dewpoint of about 60°F. This provides the moist air source for the chambers. The air from the second duct is passed through a refrigeration coil which reduces the moisture content of the air to a dewpoint of about 40°F. This provides the source of "dry" air for the chambers. A manually controlled mixing valve for each chamber mixes the proper proportions of the moist and dry air to maintain the proper RH in each chamber.

Relative humidity is measured using an EG&G Model 910 dewpoint hygrometer located in the Exposure Suite Control Center. Air from the exposure chambers is sampled from a representative location. A teflon filter is placed at the chamber end of the sample line if the test article is an aerosol. Samples of the air from each measurement location are continuously pulled through polytetrafluoroethylene (PTFE) tubing to a central location in the Exposure Suite Control Center. This assures a fresh sample at the dewpoint hygrometer. Sample air from a particular location is routed by a multiplexed valve system to either the exposure system exhaust or the dewpoint hygrometer for RH determination. The valves are controlled by either a manual switch or by a computer controlled relay, allowing RH to be measured manually or automatically by the exposure system executive computer. RH is automatically recorded at regular intervals during the 24-hour day.

Once the dewpoint has been determined by the hygrometer, the RH is automatically calculated by the exposure system executive computer using the dewpoint value and the temperature measured simultaneously at the same location by the RTD system. The following equation is used for this calculation:

$$\%RH = \frac{10 \left[9.91 - \frac{2714.55}{(5/9)(T_1 - 32) + 293.3} \right]}{10 \left[9.91 - \frac{2714.55}{(5/9)(T_2 - 32) + 293.3} \right]} \times 100$$

Where: T_1 = dewpoint temperature (°F)
 T_2 = drybulb temperature (°F)

Calibration of the dewpoint hygrometer is established prior to the start of the study and checked at least every two months following SOP # ØB-BE-3B1X. Initial calibration requires comparison at three RH levels (~ 30%, 50% and 70%) of the RH calculated by the monitor to measurements made by a calibrated portable hygrometer and RTD located near the chamber.

C. Air Flow

Air flow in the chambers is maintained by the vacuum in the central chamber exhaust duct. This vacuum is created by the chamber exhaust flow fans located in the South Equipment Room of the LSL-II Building. There are two parallel exhaust fans, one operating with the other providing backup. Both fans operate from emergency power.

Chamber air flow rate is controlled by a gate valve in each individual chamber exhaust duct. A drive motor attached to the stem of this valve allows the control of chamber flow either by computer or manually from the Exposure Control Center.

Fine control of exposure concentration can be accomplished by automatically or manually adjusting the valve position to control chamber dilution air flow within the allowable limits. Gross adjustment of concentration must be done manually by adjusting the generation system.

Chamber air flow is measured by a multiplexed orifice-meter system consisting of a calibrated orifice located in each chamber exhaust, a Validyne Model DP-45 pressure transducer, a Validyne Model CD-18 carrier demodulator, and a Validyne Model PM-12 digital voltmeter. The pressure transducer is multiplexed to each chamber's flow orifice by valves remotely controlled either manually or by means of the executive computer. This allows flow to be measured either manually or automatically. Flow is automatically recorded at regular intervals during the 24-hour day.

Calibrated flow orifice meters are located at both the inlet and exhaust to each chamber. By comparing the measured flow at the inlet and exhaust, leaks in the chamber can be detected. A leak check is automatically performed by the executive computer when each chamber is closed. If a leak is detected, the executive computer will notify the operator and will not allow exposures to proceed until the leak is repaired. This system is sensitive to very small leaks which may cause an imbalance of test article concentration within the chamber.

Calibration of the flow orifices will be done before the start of the study and at least every two months thereafter following SOP # ØB-BE-3CØV. Each flow orifice is calibrated to within ± 0.5 CFM by comparison to a calibrated digital vane anemometer temporarily inserted in the chamber duct during non-exposure periods.

D. Vacuum

The chambers are maintained at a slight negative pressure compared to the room in order to reduce the possibility of escape of test article. This negative pressure is created by the pressure drop across the HEPA and charcoal filters at the inlet to each chamber.

The same Validyne pressure transducer system used to measure chamber flows is used to measure chamber vacuum. Vacuum in the chamber is measured relative to atmospheric

pressure in the Suite Control Room. Vacuum is automatically recorded at regular intervals during the 24-hour day.

Vacuum is also continuously monitored by a mechanical pressure switch attached to each chamber. In the event of leak in the chamber, the pressure switch will immediately shut off the flow of test article to the chamber and activate an audible alarm.

V. ANIMAL FACILITY AIR HANDLING SYSTEM

Supply air enters the building through two identical parallel air handling systems (Figure 6). Each system consists of a pre-heat coil, a filter system, a heating coil, a chilling coil, and a supply fan. The pre-heat coil heats the air to a minimum of 45°F. The filter system, comprising a roll filter, pre-filter, and a bag filter, rids the air of most particles. The heating and chilling coils maintain the temperature of the air exiting the air conditioning system at about 53°F. The chilling coils also dry the air to a dewpoint not greater than 53°F.

Although simultaneous operation of both of the parallel air supply systems is necessary to provide the 20 air changes per hour typically supplied to each animal room, only one of these systems, which can be operated from the emergency power system, is required to maintain the rooms within the temperature and flow specifications required by the protocol. Exposure of the animals to the test article can continue in the event of the failure of one of the air supply systems. However, exposures generally do not continue unless all building systems are functioning normally.

The air from the two parallel building air supply systems is mixed together by an air mixing unit and is divided into two ducts which feed the rooms on the East and West sides of the animal quarters. If necessary, steam is injected into the air in these ducts to maintain the relative humidity of all rooms in the basement at a minimum of 35%. In rooms where further room RH control is necessary, it is provided by individual steam generators located in the room. Prior to entering the animal room, the air is filtered through a HEPA filter.

Air for exposure chambers is supplied to a chamber relative humidity conditioning system from the building air supply systems. A single supply system is sufficient to supply air to the RH conditioning systems for all exposure chambers in the facility.

Exhaust from the animal room is filtered by a room HEPA filter and again through a bank of building exhaust HEPA filters assuring no escape of aerosol particles from the facility. Three parallel exhaust fans provide exhaust from the rooms. Two of these fans are in operation with the third as a backup unit. One fan can be operated from the emergency power system in case of power failure.

A separate exhaust duct system for the chambers allows for the addition of scrubbers to remove particles or vapors before exhausting from the building. Because the chamber exhaust system has a separate fan, failure of one of the building exhaust fans will not prevent the continuation of exposures. However, exposures generally do not continue unless all building systems are functioning normally.

VI. EXPOSURE SUITE ALARM SYSTEM

An extensive system of alarms has been incorporated into the exposure suite automated data acquisition and control system to provide safety for the system operators, protect the health of the animals and ensure the integrity of the study. There are actually two separate

alarm systems; one provided by the computer and a separate "physical" alarm system which provides redundancy for some of the computer alarm functions.

Following each function measured by the computer, the value is compared to the alarm limit values (stored in the computer memory) for that function. There are four limit values for each function and location monitored by the computer, high and low non-critical and high and low critical. For example, chamber flows may have the following limits:

Critical low	10 air changes per hour
Non-critical Low	12
Non-critical High.....	18
Critical High	20

The result of an alarm condition depends on the function measured and the measurement location. Each function and location has an alarm response assignment in the computer. Again using chamber flow as an example, flows exceeding the non-critical limits but remaining within the critical limits will cause the computer to print a "beyond-non-critical-limits" symbol, "(" if low and ")" if high, next to the data on the daily log printed by the computer. A chamber flow which exceeds the critical limits will cause the computer to print a "beyond-critical-limits" symbol, ">" if high and "<" if low, on the daily log and to turn on the critical alarm audio alert. A critical low flow alarm will also shut off the flow of test article to the chamber. Although it is possible for the computer to make automatic corrections to air flow in the chamber, this is not done because to do so would affect concentration of the test article in the chamber. A critical low negative pressure in a chamber (which may be the result of a leak in the chamber) will also cause the computer to shut off the flow of test article to the chamber. Similar responses result from alarms arising from temperature and relative humidity measurements in the chambers, however these alarms have no effect on the operation of the test article generator.

The physical alarm system includes the following continuously monitoring devices:

- Chamber pressure (also detects critically low flow if it is the result of pump failure and not a clogged chamber inlet duct)
- Generator cabinet exhaust flow
- Chamber exhaust system flow
- Building exhaust system flow

In all cases an alarm condition from any one of these monitors is considered critical and results in the test article generator being shut off.

VII. CHAMBER EXHAUST WASTE TREATMENT

The air from the central chamber exhaust duct is mixed with the air from the entire animal facility (~ 75,000 cfm) prior to being exhausted from the building stack. Dilution of the chamber exhaust with the building exhaust results in an acceptable stack concentration usually on the order of one-tenth the Threshold Limit Value for the test article.

VIII. DATA HANDLING

Data from each exposure room are stored in the Exposure Suite Control Center on separate magnetic disks by Hewlett-Packard (HP) Model 9121 micro-floppy disc drives. Data and comments from each exposure room are printed by separate thermal dot matrix printers (HP Model 2171G) or by ink jet printers (HP Model 2225A). Data are printed and stored

immediately upon completion of the measurement to a Daily Log (example, Figure 7). Both the Daily Log and the disks will be maintained in the study files. The Daily Log will be considered the raw data. The Daily Log includes the time of measurement, the measurement location (such as chamber), the measurement function (such as temperature), the value of the measurement, the percent of target, an alarm code, and a status code. See Figure 7 for an explanation of the alarm and status codes.

At the end of the day (24-hour period), the daily data are analyzed and three summaries are printed. The first (example, Figure 8) includes the mean, % of target, standard deviation, % relative standard deviation, maximum, minimum, and number of measurements for each function (such as temperature) and location (such as chamber) monitored over the 24-hour period. The second (example, Figure 9) provides a list of outliers; that is, all data points which were beyond the defined critical operating limits. This printout allows for quick review of data which are outside the operating limits. The final summary (example, Figure 10) is a printout of all comments made by the computer, exposure specialist, and exposure operator during the 24-hour period. This includes comments on startup time, exposure termination, new calibration factors entered, and other information. This summary allows a quick review of events that occurred during the day. In addition, once each week, a chamber exposure time summary is printed (Figure 11). This summary includes total accumulated exposure time during the week for each chamber and also the mean daily length of exposure, standard deviation, maximum and minimum daily exposure time and the number of days on exposure.

Data handling and analysis procedures are described in SOPs ØB-BE-3EØB and ØB-BE-3EØE.

IX. EQUIPMENT OR POWER FAILURE PROTECTION SYSTEMS

Power is provided to the Battelle complex from two separate city substations through an automatic switching device. This significantly reduces the possibility of losing city power. Power from the city is routed to equipment in the LSL-II building through two types of motor control centers. One type can switch power from the LSL-II diesel generator. The other has access only to city power. The emergency-power-type motor control center has a low voltage detector on each leg of the three-phase input power. If the city supplied power should fail or "brown out", these detectors automatically start the emergency power diesel generator and route the emergency power to the equipment connected to the emergency-power-type motor control center.

All equipment critical to the well-being of the animals is connected to the emergency-power-type motor control center. This equipment includes:

- Emergency lighting and electrical outlets
- Building air conditioning chillers #1 and #2
- Building heating boilers and feedwater pump systems #1 and #2
- Air compressors #1 and #2
- Air supply fans #1 and #2
- Air exhaust fans #1 and #2

Note that there are two identical units of all equipment that is vital to the well-being of the animals (heating, cooling, supply air, exhaust air, and compressed air). Either of the two units has sufficient capacity to maintain the animal environment within a safe range. In all cases, the emergency power system will operate one of the two identical systems.

All building or chamber systems which are essential to the survival of the animals are alarmed. If a system malfunctions, an alarm is tripped in the Power Operator's office. A Power Operator is on duty 24 hours/day, 7 days/week. If the Power Operator is not authorized to correct the problem that caused the alarm, he immediately calls the appropriate personnel of the programs affected.

REFERENCES

- Bernstein, D.M. and Drew, R.T. (1980). "The major parameters affecting temperature inside inhalation chambers", Am. Ind. Hyg. Assoc. J., 41, pp. 420-426.
- Brown, M.G. and Moss, O.R. (1981). "An Inhalation Exposure Chamber Designed for Animal Handling", Lab. Anim. Sci., 31, pp. 717-720.
- Decker, J.R., Moss, O.R., and Kay, B.L. (1982). "Controlled-delivery vapor generator for animal exposures", Am. Ind. Hyg. Assoc. J., 43, pp. 400-402.
- Griffis, L.C., Wolff, R.K., Beethe, R.L. *et al.* (1981). "Evaluation of a Multi-Tiered Inhalation Chamber", Fundam. Appl. Toxicol., 1, pp. 8-12.
- Moss, O. R. (1980). "A chamber providing uniform concentration of particulates for exposure of animals on tiers separated by catch pans". In Workshop on Inhalation Chamber Technology, BNL 51318, Drew, R.T., editor. Brookhaven National Laboratory, Associated Universities, Inc., Upton, NY.
- Moss, O.R., Decker, J.R. and Cannon, W.C. (1982). "Aerosol Mixing in an Animal Exposure Chamber Having Three Levels of Caging with Excreta Pans", Am. Ind. Hyg. Assoc. J., 43, pp. 244-249.

Figure 1. Inhalation Exposure Chamber Designed at BNW.
Top: Oblique View of the Chamber,
Bottom: Airflow Patterns).

Figure 2. Schematic of the Chloroprene Exposure Suite.

Figure 3. Schematic of Chloroprene Vapor Generation and Distribution System.

Figure 4. Schematic of the Dewpoint and Chemical Monitoring System.

Figure 5. Schematic of Chamber Relative Humidity Control System.

Figure 6. Air Handling System for Animal Rooms of Life Sciences II Building.

Time	Data Origin	Function	Data	Target
	[LR] TFE - 625 ppm -R/M	-T90 Time Expired-Including Conc Data		
	[LR] TFE - 312 ppm -R/M	-T90 Time Expired-Including Conc Data		
	[LR] TFE - 625 ppm -R	-T90 Time Expired-Including Conc Data		
	[LR] TFE - 312 ppm -R	-T90 Time Expired-Including Conc Data		
	[LR] TFE - 156 ppm -R	-T90 Time Expired-Including Conc Data		
08:17	Hewlett Packard 85B (Access Level: Specialist)			
	TFE - 1250 ppm -R/M-Conc Data	Excluded-Data Time<T90 & Data<Target		
08:19	Hewlett Packard 85B (Access Level: Specialist)			
	TFE - 625 ppm -R/M-Conc Data	Excluded-Data Time<T90 & Data<Target		
08:21	Hewlett Packard 85B (Access Level: Specialist)			
	TFE - 312 ppm -R/M-Conc Data	Excluded-Data Time<T90 & Data<Target		
08:29	TFE - 625 ppm -R	Temperature	OKI 74.3 F	99%
08:30	TFE - 1250 ppm -R/M	Temperature	OKI 74.0 F	99%
08:32	TFE - (332) Room	Temperature	OKI 69.3 F	96%
08:34	TFE - (332) Room	Relative Humidity	OKE 53.0 %	106%
08:37	TFE - 0 ppm -R	Relative Humidity	OKI 46.0 %	84%
08:39	TFE - 0 ppm -R/M	Relative Humidity	OKI 47.0 %	85%
08:42	TFE - 156 ppm -R	Relative Humidity	OKI 48.0 %	87%
08:44	TFE - 312 ppm -R/M	Relative Humidity	OKI 49.0 %	89%
08:17	TFE - 1250 ppm -R/M	GC#34-809569	OKE 6.760E+2 ppm	55%
08:19	TFE - 625 ppm -R/M	GC#34-809569	OKE 3.445E+2 ppm	56%
08:21	TFE - 312 ppm -R/M	GC#34-809569	OKE 2.507E+2 ppm	81%
08:24	TFE - 0 ppm -R/M	GC#34-809569	OKI 0.000E+0 ppm	0%
08:26	TFE - (332) Room	GC#34-809569	OKI 0.000E+0 ppm	0%
08:29	TFE - 625 ppm -R	GC#34-809569	OKI 5.874E+2 ppm	94%
08:31	TFE - 312 ppm -R	GC#34-809569	OKI 2.854E+2 ppm	92%
08:34	TFE - 156 ppm -R	GC#34-809569	OKI 1.521E+2 ppm	98%
08:36	TFE - 0 ppm -R	GC#34-809569	OKI 0.000E+0 ppm	0%
08:40	TFE - Standard Gas-L	GC#34-809569	OKE 2.880E+1 ppm	20%
08:43	TFE - Standard Gas-H	GC#34-809569	OKI 4.993E+2 ppm	100%
08:47	TFE - 312 ppm -R	Relative Humidity	OKI 49.0 %	89%
08:49	TFE - 625 ppm -R/M	Relative Humidity	OKI 48.0 %	87%
08:52	TFE - 625 ppm -R	Relative Humidity	OKI 47.0 %	85%
08:54	TFE - 1250 ppm -R/M	Relative Humidity	OKI 53.0 %	96%
08:54	TFE - 0 ppm -R	Exhaust Air Flow	OKI 15.3 CFM	102%
08:55	TFE - 0 ppm -R/M	Exhaust Air Flow	OKI 15.0 CFM	100%
08:55	TFE - 156 ppm -R	Exhaust Air Flow	OKI 14.6 CFM	97%
08:55	TFE - 312 ppm -R/M	Exhaust Air Flow	OKI 14.6 CFM	97%
08:56	TFE - 312 ppm -R	Exhaust Air Flow	OKI 14.6 CFM	97%
08:56	TFE - 625 ppm -R/M	Exhaust Air Flow	OKI 14.9 CFM	99%
08:56	TFE - 625 ppm -R	Exhaust Air Flow	OKI 15.3 CFM	102%
08:56	TFE - 1250 ppm -R/M	Exhaust Air Flow	OKI 14.8 CFM	99%
08:45	TFE - 1250 ppm -R/M	GC#34-809569	OKI 1.212E+3 ppm	97%
08:48	TFE - 625 ppm -R/M	GC#34-809569	(OKI 5.594E+2 ppm	90%
08:50	TFE - 312 ppm -R/M	GC#34-809569	OKI 2.923E+2 ppm	94%
08:53	TFE - 0 ppm -R/M	GC#34-809569	OKI 0.000E+0 ppm	0%

Figure 7. Example of 24-Hour "Daily Log" Printout from Data Acquisition and Control Computer. See Following Page for Explanation of Columns.

Figure 7. (continued)

DESCRIPTION OF COMPUTER "DAILY LOG" OUTPUT

The date, exposure name, program version and page number will be printed at the top of each page of the daily log

- Column 1: Time -- time that measurement was taken
Column 2: Location -- location of measurement (for example, chamber)
Column 3: Function -- measurement function (for example, temperature)
Column 4: Data --

Alarm Code -- "("	Indicates data < non-critical low but $\geq$ critical low alarm value
"")	Indicates data > non-critical high but $\leq$ critical high alarm value
"<"	Indicates data < critical low alarm value
">"	Indicates data > critical high alarm value
"["	Indicates data < supercritical low alarm value
"]"	Indicates data > supercritical high alarm value

Status Code -- "OK"	Indicates monitoring instrument is functioning properly and is calibrated
"BS"	Indicates service time of monitoring instrument has expired. (Usually indicates that instrument calibration should be checked. Does not necessarily mean that data is not valid)
"I"	Indicates data will be included in summary
"E"	Indicates data will be excluded from summary
"MDM"	Indicates manual data measurement
"OED"	Indicates off exposure data

Data Value -- Data may be expressed in scientific notation (x.xxxEyy)

- Units Label -- Units of measurement (e.g., ppm, °F, mg/m³)
Column 5: Target -- % of target concentration for the measurement

Summation for the File: Nov_03_88 Exposure: Tetrafluoroethylene-Chronic

Exhaust Air Flow	Mean	% Targ	Std Dev	% RSD	Maximum	Minimum	Num	Xs
TFE - 0 ppm -R	15.5	103%	.09	1%	15.6	15.3	8.	
TFE - 0 ppm -R/M	15.1	100%	.05	0%	15.1	15.0	8.	
TFE - 156 ppm -R	14.6	98%	.03	0%	14.7	14.6	8.	
TFE - 312 ppm -R/M	14.6	98%	.03	0%	14.7	14.6	8.	
TFE - 312 ppm -R	14.7	98%	.05	0%	14.7	14.6	8.	
TFE - 625 ppm -R/M	14.9	100%	.05	0%	15.0	14.9	8.	
TFE - 625 ppm -R	15.4	103%	.06	0%	15.5	15.3	8.	
TFE - 1250 ppm -R/M	14.9	99%	.08	1%	15.0	14.8	8.	
Vacuum	Mean	% Targ	Std Dev	% RSD	Maximum	Minimum	Num	Xs
TFE - 0 ppm -R	.9	94%	.04	5%	1.0	.9	9.	
TFE - 0 ppm -R/M	1.0	98%	.02	2%	1.0	1.0	9.	
TFE - 156 ppm -R	.9	89%	.01	1%	.9	.9	9.	
TFE - 312 ppm -R/M	.9	95%	.01	1%	1.0	.9	9.	
TFE - 312 ppm -R	.8	79%	.03	4%	.8	.8	9.	
TFE - 625 ppm -R/M	.9	90%	.01	1%	.9	.9	9.	
TFE - 625 ppm -R	.9	95%	.03	3%	1.0	.9	9.	
TFE - 1250 ppm -R/M	1.1	108%	.03	3%	1.1	1.0	9.	
Relative Humidity	Mean	% Targ	Std Dev	% RSD	Maximum	Minimum	Num	Xs
TFE - 0 ppm -R	49.1	89%	3.09	6%	53.0	45.0	8.	
TFE - 0 ppm -R/M	54.6	99%	8.53	16%	65.0	44.0	8.	
TFE - 156 ppm -R	51.2	93%	3.28	6%	56.0	47.0	8.	
TFE - 312 ppm -R/M	52.9	96%	6.83	13%	60.0	43.0	8.	
TFE - 312 ppm -R	55.7	101%	4.68	8%	63.0	49.0	8.	
TFE - 625 ppm -R/M	54.5	99%	7.23	13%	62.0	43.0	8.	
TFE - 625 ppm -R	52.4	95%	4.69	9%	59.0	46.0	8.	
TFE - 1250 ppm -R/M	59.5	108%	8.25	14%	69.0	48.0	8.	
Temperature	Mean	% Targ	Std Dev	% RSD	Maximum	Minimum	Num	Xs
TFE - (336) Room	69.9	97%	.58	1%	70.6	68.9	9.	
TFE - 0 ppm -R	75.5	101%	.72	1%	76.6	74.7	9.	
TFE - 0 ppm -R/M	75.0	100%	.76	1%	76.5	73.9	9.	
TFE - 156 ppm -R	75.5	101%	.61	1%	76.3	74.7	9.	
TFE - 312 ppm -R/M	75.2	100%	.36	0%	76.0	74.7	9.	
TFE - 312 ppm -R	75.6	101%	.65	1%	76.4	74.6	9.	
TFE - 625 ppm -R/M	76.2	102%	.57	1%	77.0	75.3	9.	
TFE - 625 ppm -R	75.1	100%	.67	1%	76.0	74.3	8.	
TFE - 1250 ppm -R/M	74.6	100%	.42	1%	75.2	74.0	8.	
TFE - (332) Room	69.5	96%	.33	0%	69.9	69.0	8.	
GC#34-809569	Mean	% Targ	Std Dev	% RSD	Maximum	Minimum	Num	Xs
TFE - (332) Room	0.00E+0	0%	0.000E+0	0%	0.00E+0	0.00E+0	15.	
TFE - 0 ppm -R	0.00E+0	0%	0.000E+0	0%	0.00E+0	0.00E+0	15.	
TFE - 0 ppm -R/M	0.00E+0	0%	0.000E+0	0%	0.00E+0	0.00E+0	15.	
TFE - 156 ppm -R	1.53E+2	98%	2.010E+0	1%	1.56E+2	1.49E+2	13.	
TFE - 312 ppm -R/M	3.07E+2	98%	5.478E+0	2%	3.13E+2	2.92E+2	12.	
TFE - 312 ppm -R	3.09E+2	99%	1.068E+1	3%	3.21E+2	2.85E+2	13.	

Figure 8. Example of 24-Hour "Data Summation" Printout from Data Acquisition and Control Computer.

Outlier Table for the File: Nov_21_88 Exposure: Acetonitrile

Origin	Instrument	Time	Data	Lower	Target	Higher	
Aceto -	0 ppm-M	Relative Humidity	10:14	33.0	35.0	55.0	75.0

Figure 9. Example of 24-Hour "Data Outlier Table" Printout from Data Acquisition and Control Computer. Table shows data which were beyond the defined Critical Limits.

Daily Comments		Tetrafluoroethylene-Chronic	File: Nov_03_88
Time	Operator	Comment	
07:51		Chamber Leak Check for TFE - 156 ppm -R	
07:51		Exhaust Flow= 14.6 Inlet Flow= 15.1 (-3.4% leak) [Acceptable]	
07:52		Chamber Leak Check for TFE - 0 ppm -R	
07:52		Exhaust Flow= 15.2 Inlet Flow= 15.6 (-2.6% leak) [Acceptable]	
07:52		All Chambers have been found ACCEPTABLE.	
08:01	Hewlett Packard 858	TFE - Standard Gas-M-Conc Data Excluded-Exposure Not Running.	
08:04		HP858 found Parameters Okay & Ready for Exposure Start.	
08:14	Hewlett Packard 9816	Exposure Timing started. [Time=T(0)]	
08:15		TFE Chronic - Main Distribution Valve - Valve Opened	
08:15		[LR] TFE Chronic - 1250 ppm Chamber Rats/Mice - Valve Opened	
08:15		[LR] TFE - 1250 ppm -R/M -ON Exposure-Enable Environ Data Collection	
08:15		[LR] TFE Chronic - 625 ppm Chamber Rats/Mice - Valve Opened	
08:15		[LR] TFE - 625 ppm -R/M -ON Exposure-Enable Environ Data Collection	
08:15		[LR] TFE Chronic - 312 ppm Chamber Rats/Mice - Valve Opened	
08:15		[LR] TFE - 312 ppm -R/M -ON Exposure-Enable Environ Data Collection	
08:15		[LR] TFE Chronic - 625 ppm Chamber Rats - Valve Opened	
08:15		[LR] TFE - 625 ppm -R -ON Exposure-Enable Environ Data Collection	
08:15		[LR] TFE Chronic - 312 ppm Chamber Rats - Valve Opened	
08:15		[LR] TFE - 312 ppm -R -ON Exposure-Enable Environ Data Collection	
08:15		[LR] TFE Chronic - 156 ppm Chamber Rats - Valve Opened	
08:15		[LR] TFE - 156 ppm -R -ON Exposure-Enable Environ Data Collection	
08:15		[LR] TFE Chronic - Standard Gas Valve - Valve Opened	
08:27		[LR] TFE - 1250 ppm -R/M -T90 Time Expired-Including Conc Data	
08:27		[LR] TFE - 625 ppm -R/M -T90 Time Expired-Including Conc Data	
08:27		[LR] TFE - 312 ppm -R/M -T90 Time Expired-Including Conc Data	
08:27		[LR] TFE - 625 ppm -R -T90 Time Expired-Including Conc Data	
08:28		[LR] TFE - 312 ppm -R -T90 Time Expired-Including Conc Data	
08:28		[LR] TFE - 156 ppm -R -T90 Time Expired-Including Conc Data	
08:17	Hewlett Packard 858	TFE - 1250 ppm -R/M-Conc Data Excluded-Data Time<T90 & Data<Target	
08:18		TFE - 625 ppm -R/M-Conc Data Excluded-Data Time<T90 & Data<Target	
08:21		TFE - 312 ppm -R/M-Conc Data Excluded-Data Time<T90 & Data<Target	
10:25	Gary R. Eli	Service status updated on GC concentration from data collected	
10:25		11-02-88 gas bag samples.No correction needed,as per Mr.	
10:25		Reassign.	
10:26		Concentration Monitor-Service/Status	
10:26		[1. 1]TFE - 1250 ppm -R/M Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 2]TFE - 625 ppm -R/M Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 3]TFE - 312 ppm -R/M Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 4]TFE - 0 ppm -R/M Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 5]TFE - (332) Room Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 6]TFE - 625 ppm -R Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 7]TFE - 312 ppm -R Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 8]TFE - 156 ppm -R Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1. 9]TFE - 0 ppm -R Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
10:26		[1.12]TFE - Standard Gas-M Srv Date was: 27 Oct 1988 ts: 3 Nov 1988	
14:27	Hewlett Packard 9816	[LR] TFE Chronic - Main Distribution Valve - Valve Closed	
14:27		Exposure Terminated. Exposure timers stopped.	
14:27		Generator valve already OFF.	
14:27		[LR] TFE Chronic - 1250 ppm Chamber Rats/Mice - Valve Closed	
14:27		[LR] TFE - 1250 ppm -R/M -OFF Exposure-Enable Environ Data Collection	

Figure 10. Example of 24-Hour "Comment Summary" Printout from Data Acquisition and Control Computer. Table shows a summary of all comments recorded on "Daily Log" printout.

Chamber Exposure Time Summary							Acetonitrile
From: November 15, 1988 To: November 21, 1988							
Chamber Name		Total	Average	Std Dev	Maximum	Minimum	# Days
Aceto - 400 ppm-R		30:01	06:00	00:26	06:01	06:00	5
Aceto - 200 ppm-M		30:00	06:00	00:00	06:00	06:00	5
Aceto - 200 ppm-R		30:00	06:00	00:00	06:00	06:00	5
Aceto - 100 ppm-M		30:00	06:00	00:00	06:00	06:00	5
Aceto - 100 ppm-R		30:00	06:00	00:00	06:00	06:00	5
Aceto - 50 ppm-M		30:01	06:00	00:26	06:01	06:00	5
Aceto - 0 ppm-R							
Aceto - 0 ppm-M							

Standard Deviation is in Minutes & Seconds. All Others are in Hours & Minutes
Exposure Times DO NOT include T90 time.

Figure 11. Example of Weekly Chamber Exposure Time Summary.

CHLOROPRENE
RABBIT DEVELOPMENTAL TOXICITY

STUDY PROTOCOL

ØB-DT-1F36-ØØ-Ø27Ø
ATTACHMENT III

STANDARD OPERATING PROCEDURE:

ARTIFICIAL INSEMINATION OF RABBITS

STANDARD OPERATING PROCEDURE

Document Code

OB-DT-3B0E-03

Date of Implementation

2/28/90

Page 1

Title: Artificial Insemination of Rabbits

Originated by R. L. Rommerson Date 2/28/90

Approved by Judy G. Mast Date 3/6/90
Task Leader

Approved by Betty J. Cho Date 3/6/90
Technical Group Manager

Approved by _____ Date ____/____/____
(Title : _____)

Circulation List:

Taskleader (2)
QAU

Record of Revisions, Changes, and Reviews:

Rev Lener	Date Implemented	Comment	Signature/Date	
			Task Leader	Tech Grp. Mgr.

A. Assemble materials for artificial insemination.

1. Semen Collection

- a. Artificial vaginas (AVs); see Section B for assembly instructions.
- b. K-Y jelly
- c. Thermometers - calibrated
- d. Sink with hot running water
- e. Heavy rubber glove with long (elbow length) arm and cut-off fingers

2. Sperm Evaluation

- a. Microscope
- b. Calculator
- c. Slides and cover slips
- d. Pasteur pipettes and bulbs
- e. RBC diluting pipettes
- f. Disposable pipettes 1, 5, and 10 ml
- g. Saline (0.9%)
- h. 1% formal saline (1 ml formalin in 100 ml saline)
- j. Hemacytometer
- k. Beakers and Haemo-sol (for cleaning hemacytometer)
- l. Hand counter

3. Chorionic Gonadotropin Injection

- a. Gauze squares
- b. Sterile saline
- c. Chorionic gonadotropin, 500 USP units/ml in saline (use 0.2 ml/dose--100 units/doe) obtained at a local pharmacy by prescription from our staff veterinarian.
- d. 1 or 2 ml syringes with 22-G needles

4. Insemination

- a. Semen extender; see section E
- b. Insemination pipettes (plastic or glass tubing, 3-7 mm diameter with a smooth rounded end) attached to a syringe
- c. 50 ml Plastic centrifuge tube with cap for inseminant

B. Preparation of AVs (See Figure 1.)

1. Materials for AV

- a. Smooth, flexible radiator hose (4 cm diameter x 8 cm long)
- b. Latex AV collection funnel (Nasco; Modesto, California), shortened to ~15 cm
- c. Wide rubber band (can be cut from top of the AV liner)
- d. Rubber stopper, size 9, with 2 holes
 - (1) 1.5 cm hole to hold semen collection tube
 - (2) 0.5 cm hole for glass tube
- e. Glass tube (0.5 cm diameter x 6 cm long) with fire-polished ends
- f. Tygon tubing (0.4 cm diameter x 5 to 6 cm long)
- g. Pinch clamp for tubing
- h. Semen collection tube; graduated centrifuge tube (plastic, 15-20 ml volume cut down to 5 ml volume, 1.5 cm diameter with polished edges)

2. Assembly of AV

- a. Insert glass tubing into the small hole of the rubber stopper until flush with the bottom of the stopper.
- b. Fit tygon tubing over end of exposed glass tubing.
- c. Insert the AV liner into radiator hose, leaving approximately 4 cm of the top of the liner exposed.
- d. Fold top of the liner over radiator hose and secure with the wide rubber band.
- e. Insert narrow end of the AV liner through the large hole of the rubber stopper, making sure the stopper will plug into the radiator hose.
- f. Insert the semen collection tube into the narrow end of the AV liner, covering the open end of the tube enough to create a good seal. Push the tube up into the stopper securely.
- g. Place clamp on Tygon tubing.

FIGURE 1. Materials and Preparation of Artificial Vagina for Rabbit Semen Collection

3. Preparation of AV for Semen Collection

- a. Remove the stopper and fill assembled AV with hot tap water. Stopper the radiator hose tightly to create a closed-system.
NOTE: Avoid introduction of water into AV lining or collection tube as water will kill sperm.
- b. Hold the AV upside down with the Tygon tubing toward your mouth. Open the clamp and blow into the tubing to inflate the AV. Reclamp while inflating.
- c. Insert the thermometer ~3 cm into the end of the AV to check the temperature, 42-45°C. If the temperature is too high, the male will urinate. If it is too low, the male will not ejaculate.
- d. Form the folds of the liner into a "T" shape if possible. Smear the inflated end of the AV liner with K-Y jelly.
- e. Prepare a sufficient number of AVs to attempt semen collection from all of the bucks.

4. Cleaning of the AV

- a. The AV must be cleaned after each ejaculate. Clean the AVs after all collections are made.
- b. Detach the narrow end of the liner from the stopper and invert the liner. Wash all parts thoroughly with a detergent solution (Haemo-sol), rinse with water, then with 70% alcohol, and allow to air dry.

C. Semen Collection

The bucks must be trained during the quarantine period (~4 weeks) to serve an AV with the assistance of a teaser doe. This training period also tends to improve semen quality. Semen collection may be attempted daily; obtain approximately 2-3 samples per buck per week.

1. Put on the heavy, elbow length rubber glove to protect your arm from the rabbit's claws.
2. Carry the teaser doe and the prepared AV to the buck's cage. Do not place the buck in the female's cage as this is not his territory.
3. Hold the teaser doe securely behind her head with one hand, position her over your other arm with her hind legs straddling your hand and the AV. Hold the doe's tail down over the vaginal area to help protect her from the male.

4. Position the opening of the AV just under the teaser doe's tail. Allow the male to mount the teaser doe and, if necessary, slowly move the AV towards the penis. Caution should be used to avoid allowing the male to inseminate the teaser doe.
5. After ejaculation into the AV, return the doe to her cage and visually check the contents of the collection tube. Discard the sample if any urine (yellow sediment) or blood is observed. A clear, gelatinous material may be seen. This is the copulation plug and does not interfere with the purity of the sample. Healthy semen is a chalky white color.
6. To remove the collection tube with the pure semen sample, carefully empty the water out of the liner jacket and pull the tube gently from the stopper, making sure the liner is still securely attached. Dry the tube and liner with a Teri towel and gently roll the liner away from the top of the tube with your thumb while holding the tube in the same hand. DO NOT allow any moisture to come into contact with the collection tube contents as water will kill the sperm. Cover the collection tube with parafilm and place in numbered slot in the holding tray. Record the buck number corresponding to the sample number.

D. Semen Evaluation (see form on page 11)

1. Motility Estimation

- a. Remove the copulation plug (if present) with forceps. Place a drop of seminal fluid and a drop of saline on a slide and cover with cover slip.
- b. Examine sperm for active wave motion and a large number of active sperm. Record the following:
 - (1) date
 - (2) buck number
 - (3) volume of ejaculate minus the copulation plug
 - (4) % motility and motility score 0 to 4 (4 is best)
 - (5) technician ID

2. Pooling Samples

Pool a minimum of 3 acceptable samples into a graduated plastic centrifuge tube (50 ml). Use samples with $\geq 70\%$ motility and a score of ≥ 3 .

3. Sperm Count

- a. Mix the pooled sample by gentle swirling.
- b. Fill the RBC pipette to the 0.5 mark with semen.
- c. Wipe the pipette clean and immediately fill to the 101 mark with 1% formal saline (This is a 1:200 dilution.). Place one finger over the tip of pipette and remove the suction apparatus from the opposite end. Place your thumb and forefinger over each end. Shake the pipette vigorously to mix semen and formal saline (~30 seconds).
- d. Discharge and discard fluid from the pipette stem and part of the bulb (approximately one-third of the bulb contents).
- e. Charge both chambers of the hemacytometer with the fluid remaining in the RBC pipette and let it stand briefly.
- f. Use a 10X eyepiece to read the hemacytometer. Count the number of sperm in the 1-mm squares (large center square and four corners) until the count is greater than 200 sperm. Count up to five 1-mm squares. Sperm lying on the lines on the bottom and right sides of the large squares are included in the count, but not those on the top or left lines. Make two counts, one from each chamber. Record the average.
- g. Clean the hemacytometer by immersing in hot Haemo-sol solution. Gently rub the hemacytometer and cover slip with fingers and rinse both in hot water then 70% ethanol. Wipe dry with a gauze pad; DO NOT drain dry. Flush RBC pipettes following the same procedure, then air dry. A drying oven may be used.

4. Calculation of Sperm Count

- a. The sperm count equation is:

$$\text{Sperm/ml} = \text{number sperm counted} \times \text{Dilution Factor}$$

$$\text{Dilution Factor} = \frac{200^*}{[\text{Number of 1-mm Squares Counted}][1 \times 10^{-4} \text{ ml/square}]^{**}}$$

*200 = dilution from RBC pipette.

**Volume of a 1-mm square.

Calculated Dilution Factors are listed below:

<u>Number of 1-mm Squares Counted</u>	<u>Dilution Factor</u>
1	2 x 10 ⁶
2	1 x 10 ⁶
3	0.66 x 10 ⁶
4	0.5 x 10 ⁶
5	0.4 x 10 ⁶

b. Example:

If 2 1-mm squares had a count of 284 sperm

$$\text{Sperm/ml} = (284) (1 \times 10^6) = 284 \times 10^6 \text{ sperm/ml}$$

5. Calculation for Semen Extension

a. Required: 0.5 ml inseminant/doe for AI, 20-30 x 10⁶ sperm/ml

The term inseminant is used for the mixture of semen plus extender.

b. Total number of sperm in the pooled sample is calculated as follows:

$$\text{Total sperm} = (\text{sperm/ml pooled semen}) \times (\text{ml pooled semen})$$

c. Calculate the dilution necessary to yield 20-30 x 10⁶ sperm/ml in the final volume of inseminant. Allow at least 1-2 ml extra volume for losses during insemination.

$$\text{ml inseminant} = \frac{\text{total sperm}}{20-30 \times 10^6 \text{ sperm/ml}}$$

Example:

Given 284 x 10⁶ sperm/ml and 1.8 ml pooled semen

$$\text{Total sperm} = (284 \times 10^6 \text{ sperm/ml}) (1.8 \text{ ml pooled semen}) = 511 \times 10^6 \text{ total sperm}$$

$$\text{ml inseminant} = \frac{511 \times 10^6 \text{ Sperm}}{25 \times 10^6 \text{ sperm/ml}^*} = 20.4 \text{ ml Inseminant}$$

*25 x 10⁶ sperm/ml inseminant is within the range of 20 - 30 x 10⁶ required.

20 ml of inseminant is sufficient to breed 36 does plus have approximately 2 ml for volume loss and post-insemination examination.

- d. Perform dilution using extender at room temperature. Cap the tube and mix contents by gentle inversion.
- e. Store the tube containing the extended semen at approximately 35°C throughout the insemination procedures.

E. Preparation of Semen Extender

One of the following extenders may be used.

1. Oviduct Stimulating Medium (OSM)

This semen extender can be prepared up to one week prior to use and refrigerated until used. The following chemicals are added per liter of boiled, cooled distilled water:

NaCl	6.30 g
KCl	.75 g
MgCl ₂	.10 g
CaCl ₂	.37 g
NaHCO ₃	3.36 g
NaHPO ₄	.069 g
Glucose	.270 g
EDTA (sodium)	.15 g
Penicillin/streptomycin (10,000 IU/10 mg)	6 ml

Adjust the pH to 7.4 each day the OSM is to be used.

2. Buffered egg yolk extender may be prepared, one to two days prior to AI:

- a. Clean and check all supplies.
- b. Obtain a supply of fresh eggs.
- c. Prepare TRIS-citrate buffer for extender.

(1) Weigh the following:

- 3.028 g TRIS (hydroxymethylaminomethane)
- 1.675 g citric acid
- 1.250 g glucose

(2) Place chemicals in a 100-ml volumetric flask, dissolve, and make up to volume with boiled and cooled distilled water.

(3) Label flask and store at room temperature.

- e. On each day of AI prepare semen extender. Mix 6 ml egg yolk and 30 ml TRIS-citrate buffer. Adjust pH to 6.8 to 6.9 with 2.5 N NaOH or dry citric acid.

F. Preparation of Chorionic Gonadotropin for Injection

- 1. Dilute the contents of the vial with sterile saline according to package instructions. Do not use the diluent that comes with the chorionic gonadotropin.
- 2. Shave both ears of all breeding does 1 to 2 days before insemination. See Section G.7 for injection procedures.

G. Artificial Insemination (AI)

- 1. Begin insemination within 2 hours of semen collection.
- 2. Mix the inseminant by gentle swirling of the tube. Draw 0.5 ml into the insemination pipette. Draw air into syringe prior to drawing up fluid to allow space for delivery of the total volume. A new pipette is used for each doe.
- 3. Coat the end of pipette with K-Y jelly.
- 4. An assistant will position the doe for insemination as follows: The assistant will be seated. Place the doe on the floor, facing the assistant between his/her feet, and lift the doe's rear quarters between the assistant's knee so that the doe's head is near the floor and her back is toward the assistant. Hold the doe's hind feet and legs securely. Lateral support is provided by the assistants knees and legs.
- 5. Remove any fecal material present with a gauze pad. Insert the end of the pipette into the vagina and rotate with gentle pressure until it slides under the pubic bone and up to the cervix.
- 6. Discharge the pipette and withdraw it from the vagina.
- 7. After insemination, inject the doe in a marginal ear vein with 0.2 ml of the chorionic gonadotropin. This causes the ovaries to ovulate.
- 8. Patting the doe on the bottom as she is returned to her cage may improve ovulation and pregnancy rate.
- 9. If sufficient inseminant is left after the insemination procedure, evaluate sperm motility and % motile.

RABBIT INSEMINATION RECORD

ØB-DT-3BØE-Ø3

Page 11 of 11

PROJECT: NTP-IRT
STUDY: RABBIT TERATOLOGY
TECH ID: _____

CHEMICAL: CHLOROPRENE
INVESTIGATOR: TJ MAST
DATE: _____

A. Semen Collection: Time _____

<u>Buck #</u>	<u>Sample #</u>	<u>Motility(0-4)</u>	<u>%Motility</u>	<u>Volume (ml)</u>	<u>Comments</u>
_____	_____	_____	_____	_____	_____
_____	_____	_____	_____	_____	_____
_____	_____	_____	_____	_____	_____
_____	_____	_____	_____	_____	_____
_____	_____	_____	_____	_____	_____

Pooled Volume: _____

B. Sperm Count: Time _____

<u>#1-mm Squares</u>	<u>Dilution Factor x10⁶</u>	<u>Sperm Count</u>	<u>Pooled Volume (ml)</u>	<u>Total Sperm x10⁶</u>
1	2	x _____	x _____	= _____
2	1	x _____	x _____	= _____
3	0.66	x _____	x _____	= _____
4	0.50	x _____	x _____	= _____
5	0.40	x _____	x _____	= _____

Note: Total sperm must be at least _____ x10⁶ for today's insemination based on

_____ does and 20 x 10⁶ sperm/ml(minimum) and _____ ml inseminant.

_____ does x 0.5 ml inseminant/doe = _____ ml inseminant needed.

Addition of _____ ml extra makes the total volume prepared today = _____ ml inseminant

C. Semen Extension

_____ ml inseminant = _____ x10⁶ total sperm + _____ x10⁶ sperm/ml

Note: Sperm/ml of inseminant must be 20 - 30 x10⁶

_____ ml pooled semen + _____ ml extender = _____ ml inseminant

D. Chorionic Gonadotropin: 5000 units/10ml sterile saline

Lot # _____ Expiration date _____

Dosage: 0.2 ml/doe (100 units/doe)

Note: Up to 50 does can be injected from one 10 ml bottle.

E. Insemination: Start Time _____ Completion Time _____

F. Post-insemination Sperm Examination: Time _____

Sperm motility (0-4) _____ % Motility _____

Cage Maps

PROJECT: NTP-IRT
STUDY: RABBIT TERATOLOGY
ROOM: 432
Acclimation Grp A only

CHEMICAL: CHLOROPRENE
INVESTIGATOR: T. MAST
DATE: 3/23/90 - 3/25/90
FRIDAY - SUNDAY

CHAMBERS: CONTROL

7

	LEVEL 1
LEVEL 2	
	LEVEL 3
LEVEL 4	

8

	LEVEL 1
	6
LEVEL 2	9
14	
15	LEVEL 3
	16
LEVEL 4	

COMMENTS:

CHAMBERS: 10 PPM

1

	LEVEL 1
	4
LEVEL 2	5
13	
19	LEVEL 3
LEVEL 4	

2

	LEVEL 1
LEVEL 2	
	LEVEL 3
LEVEL 4	

COMMENTS:

VERIFIED BY: RRR JJE

PROJECT: NTP-IRT
STUDY: RABBIT TERATOLOGY
ROOM: 432

CHEMICAL: CHLOROPRENE
INVESTIGATOR: T. MAST
DATE: 3/23/90 - 3/25/90

Acclimation Grp 1 only

CHAMBERS: 40 PPM

3

	LEVEL 1
	2
LEVEL 2	7
10	
12	LEVEL 3
LEVEL 4	

4

	LEVEL 1
LEVEL 2	
	LEVEL 3
LEVEL 4	

COMMENTS:

CHAMBERS: 175 PPM

5

	LEVEL 1
	1
LEVEL 2	3
8	
18	LEVEL 3
LEVEL 4	

6

	LEVEL 1
LEVEL 2	
	LEVEL 3
LEVEL 4	

COMMENTS:

VERIFIED BY:

RRR

APPENDIX E

QUALITY ASSURANCE

Quality Assurance Statement

**INHALATION DEVELOPMENTAL TOXICITY STUDY OF
CHLOROPRENE IN RABBITS**

Quality Assurance Statement

Listed below are the phases and/or procedures included in the study described in this report which were reviewed by the Quality Assurance Unit during the period 1/1/90 - 4/30/90, or specifically for this study. Included are the dates the reviews were performed and findings reported to management. (Any findings likely to affect the outcome of the study were reported to the study director or his designee at the time of the review.)

Phase/Procedure Reviewed	Review Date	Date Findings Submitted in Writing to Study Director/Management
Animal Receipt	1/11/90	1/18/90
Body Weights	2/13/90	2/20/90
Clinical Observations	3/5/90	3/5/90
Dosing	4/3/90*	4/13/90
Necropsy	4/19/90*	5/11/90
Teratological Examinations	4/19/90*	5/11/90
Data	7/9-10/90*	8/8/90
Draft Report	8/3,6,9,10,13,14/90	8/15/90
Final Report	4/21,25,26/94	4/26/94

*Reviewed specifically for this study

Quality Assurance Auditor

5/11/94
Date

DISTRIBUTION

No. of
Copies

OFFSITE

2 DOE/Office of Scientific and Technical Information

10 Dr John Bucher
NIEHS, NTP, DTRT
P.O. Box 12233
Research Triangle Park, NC 27709

Elizabeth Ford
NIEHS, CPMB
P.O. Box 12874
Research Triangle Park, NC 27709

ONSITE

DOE Richland Operations Office

D.K. Jones

Pacific Northwest Laboratory

14 T.J. Mast (5)
R.A. Gelman
Program Office (2)
Publishing Coordination
Technical Report Files (5)

DATE

FILMED

6/27/94

END