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UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

NOV 13 1996

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OFFICE OF
PREVENTION, PESTICIDES, AND
TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Chloropicrin (CP): Mouse oncogenicity (MRID 43632201) and rat oncogenicity (MRID No. 43755301) studies. D223393; D215526 and D222580.
Chemical ID #081501
Chemical No. 214

TO: Paula Deschamp, PhD.
Registration Coordination Analysis Branch
Health Effects Division (7509C)

FROM: Stanley B. Gross, PhD, DABT, CIH
Toxicologist/Hygienist
Toxicology Branch I
Health Effects Division (7509C)
10/7/96

THRU: Joycelyn E. Stewart, PhD
Head, Section II, Toxicology Branch I
Health Effects Division (7509C)
10/30/96

cc: Larry Schnaubelt/Susan Jennings (PM 72)
Reregistration Review Branch
Registration Division (7508W)
KB 11/11/96

I. SUBMISSION/REQUEST.

The Registrant (The Chlorpicrin Manufacturers Task Force c/o Nicklor Chemical Co., Long Beach CA) has submitted OPP the following oncogenicity studies for review:

1) Rat Oncogenicity Study (OPPTS 870.4100 §83-1): Burleigh-Flayer, H.D. and Benson, C.L. (1995) Chloropicrin: Vapor Inhalation Oncogenicity Study in CD Rats. Bushy Run Research Center (BBRC), Union Carbide Corporation, Export PA; Study No. 92N1106 (MRID No. 43755301), July 29, 1995, Unpublished.

2) Mouse Oncongencidity study (OPPTS 870.4100 §83-1b: Burleigh-Flayer, W.J. Kintigh, and C.L. Benson (1995) Chloropicrin: Vapor Inhalation Oncogenicity Study in CD-1® Mice (MRID No.43632201); Bushy Run Research Center, Export PA 15632. Laboratory project No. 92N1105; April 25, 1995; Unpublished; and pathology peer review:

Robert M. Kovatch, 1996). Pathology Peer Review of

Chloropicrin: Vapor Inhalation Oncogenicity Study in CD-1(R) Mice. Study No. 92N1105. Pathology Associates International Corporation, Frederick, MD. January 31, 1996. Unpublished.

II. CONCLUSIONS:

The DERs for the submitted studies are attached. Summaries are included below. Both studies are ACCEPTABLE and meet the guideline requirements for mouse and rat inhalation oncogenicity studies.

III. STUDY SUMMARIES.

A. Rat Oncogenicity Summary:

In an inhalation oncogenicity study (MRID 43755301), groups of 50 male and 50 female Sprague-Dawley CD rats were exposed (whole body) to chloropicrin vapor (99.6% pure) for up to 108 weeks at concentrations of 0, 0.1, 0.5, or 1.0 ppm. Toxicity was monitored by clinical observations, body weights and gains, food consumption, ophthalmology examinations, hematology, organ weights, necropsy observations and microscopic evaluation.

In the 0.5 and 1.0 ppm groups, there was a dose-dependent decrease in survival time and an increase in mortality rate in males. Survival at week 108 was 34% and 30% in males exposed at 0.5 or 1.0 ppm compared to 58% in controls and mean survival times were 672 and 647 days compared to 696 days for controls ($p<0.05$ and 0.01). Based on mortality and longevity, females appeared to be more tolerant than males in this study, an observation consistent with a previously submitted 90 day inhalation study.

Minimal but significant effects on body weights and gains were noted for both sexes but only in the first 2 weeks of the study. Males exposed at 1.0 ppm, had an increased incidence of rhinitis in the anterior nasal cavity (70% vs 40% in controls). There were no statistical increases in neoplasm incidence at any site and the incidence of neoplasms in all groups of males and females was similar to that normally seen in rats of this strain. However, one male and one female from the high dose groups were found to have a mast cell sarcoma of the larynx, which was cited by the reviewing pathologist as being a very rare tumor in rats. The significance of this tumor was considered uncertain.

The systemic LOEL is 0.5 ppm, based on decreased survival (males) and marginally decreased body weight gains in the first few weeks of the study (both sexes). The NOEL for males was 0.1 ppm and 1.0 ppm for females. Under the conditions of the study there was no oncogenic response. There were no increases in neoplasm incidence at any site and the incidence of neoplasms in all groups of males and females was similar to that normally seen in rats of this strain.

Dr. Lucas H. Brennecke, HED's consulting pathologist, was asked to comment on the significance of the mast cell sarcomas seen in the high dose male and high dose female. Dr. Brenneke's recommendations (25 October 1996) are attached to this memorandum.

This chronic toxicity study in the rat is acceptable and does satisfy the guideline requirement for a chronic inhalation study (83-2a).

COMPLIANCE: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

B. Mouse Oncogenicity Summary.

In a vapor inhalation oncogenicity study (MRID 43632201), groups of CD-1® mice (50 mice/sex) were exposed to concentrations of 0 (air), 0.10, 0.50 or 1.0 ppm chloropicrin vapor (99.6% pure) for 6 hours/day, 5 consecutive days/week for 78 weeks. Toxicity was monitored by clinical observations, body weights and gains, food consumption, ophthalmology examinations, hematology, organ weights, necropsy observations and microscopic evaluation.

There were no toxic signs or mortality associated with the CP exposures. In males and females exposed to 0.5 ppm, mean body weights were lower and cumulative weight gains were depressed throughout the study. At the 1-ppm exposure level, mean body weights/weight gains at week 27 were significantly ($p<0.01$) decreased (5%/19% in males and 9%/32% in females). Food consumption was decreased in females. At week 27, mean weights were decreased 5% and 9% as compared to controls in males and females,respectively, and weight gains were 13% and 24% lower. Food consumption in females was 5% decreased (weeks 1-13). At sacrifice, lung weights were increased 12% (males) and 17% (females). In the nasal cavity, rhinitis with serous exudate was significantly increased in both sexes and hyaline epithelial inclusions and olfactory epithelial atrophy were significantly increased in females.

Non-neoplastic lesions related to dosing with CP related to the lungs and the nasal cavity. Lung bronchiectasis characterized by dilation of the bronchus was accompanied by thickened bronchial wall (fibrosis) and minimal inflammatory exudates elevated in both sexes exposed to 0.5 and 1.0 ppm.
In the nasal cavity, serous exudate and atrophy of the olfactory epithelium, decreased olfactory neurons were increased in both sexes exposed to 0.5 and 1.0 ppm. The lesions of the nasal cavity seen at 0.5 ppm were increased in incidence in both sexes and olfactory epithelial atrophy was present in 72-80% of the mice. In addition to the lung lesions at 0.5 ppm, alveolar histiocytosis was observed in 29/50 males and 35/50 females ($p<0.01$) and alveolar/ bronchiolar cell hyperplasia was present in 6/50 1-ppm females ($p<0.05$).

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Neoplastic findings -- incidence of pulmonary edema were slightly increased but not significant statistically, in male and female mice of the 0.5 and 1.0 ppm groups. The slightly elevated indices in 0.5 and 1.0 ppm male and female groups were attributed to random variation.

The LOEL for systemic toxicity and irritation is 0.5 ppm based on lower body weight and body weight gains, increases in lung weight and histologic changes in the nasal cavity, upper respiratory tract and lungs. The systemic NOEL is 0.1 ppm. CP was not neoplastic in this study.

This oncogenicity study in the mouse is acceptable and satisfies the guideline requirement for an oncogenicity study in mice (83-2b).

cponcos.m96 E1 October 31, 1996

A

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A Company of Science Applications International Corporation
SAIC
An Employee-Owned Company

MEMORANDUM

SUBJECT: Chloropicrin (CP): Pathology consult for rat oncogenicity study
MRID No. 43755301
Chemical ID #081501
Chemical No. 214

TO: Stanley B. Gross, PhD, DABT, CIH
Toxicologist/Hygienist
Toxicology Branch I
Health Effects Division (7509C)

FROM: Lucas H. Brennecke, DVM, DACVP
Pathology Consultant
Pathology Associates International

THRU: Esther Rinde, Ph.D.
Carcinogenicity Peer Review Manager
Science Analysis Branch
Health Effects Division (7509C)
US Environmental Protection Agency
401 M St. NW
Washington, DC 20460

DATE: 25 October 1996

Action Requested: Provide recommendations concerning the significance of mast cell sarcomas seen in one male and one female of the high dose groups

I completely agree with the following comments which were provided in the pathology report (P. 9 of Appendix 2), "Mast cell sarcomas are very rare tumors in rats at any site. They are common skin tumors in dogs and cats, but would be unusual, even in these species, in the larynx." Furthermore, I could find no report of any mast cell tumors having been reported in the larynx of rats.

For the purposes of scientific interest, it might be valuable to have the tumors reviewed by another pathologist and/or perform special histochemical or immunohistochemical staining to confirm the diagnoses of mast cell sarcomas in the two rats. Additionally, transmission electron microscopy could be conducted to confirm the diagnoses. The fact remains, however, that only one such tumor in each sex was noted, and there were no other statistically significant treatment-related increases in neoplasms in either sex.

It is my opinion that the tumors may be mis-diagnoses, but even if the diagnoses were confirmed, their significance would be marginal at best.

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DATA EVALUATION REPORT

012099

CHLOROPICRIN

Study Type: 83-2a; Inhalation Oncogenicity Study in Rats

Dynamac Study No. 123A (MRID No. 43755301)

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
1921 Jefferson Davis Highway
Arlington, VA 22202

Prepared by

Pesticides Health Effects Group
Sciences Division
Dynamac Corporation
2275 Research Boulevard
Rockville, MD 20850-3268

Primary Reviewer:
William L. McLellan, Ph.D

Signature: William L. McLellan
Date: 4/15/96

Secondary Reviewer:
Sandra Daussin

Signature: Sandra Daussin
Date: 4/19/96

Project Manager:
William J. Spangler, Ph.D.

Signature: William J. Spangler
Date: 05/15/96

Quality Assurance:
Reto Engler, Ph.D.

Signature: Reto Engler
Date: 05/14/96

Disclaimer

This Data Evaluation Report may have been altered by the Health Effects Division subsequent to signing by Dynamac Corporation personnel.

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CHLOROPICRIN Inhalation Oncogenicity Study-Rat (83-2)

EPA Reviewer: Stanley Gross, Ph.D. _____, Date 10/7/96
Review Section II, Toxicology Branch I (7509C)
EPA Secondary Reviewer: Marion Copley, D.V.M. _____, Date 11/13/96
Review Section IV Toxicology Branch I (7509C) copy

DATA EVALUATION RECORD

STUDY TYPE: Chronic Inhalation Oncogenicity OPPTS 870.4100 §83-2a

DP BARCODE: 219429 SUBMISSION CODE: S494198
P.C. CODE: 081501 TOX. CHEM. NO.: 214

TEST MATERIAL (PURITY): Chloropicrin (99.6%)

SYNONYMS: Nitrochloroform, trichloronitromethane

CITATION: Burleigh-Flayer, H.D. and Benson, C.L. (1995) Chloropicrin: Vapor Inhalation Oncogenicity Study in CD Rats. Bushy Run Research Center (BBRC), Union Carbide Corporation, Export PA; Study No. 92N1106 (MRID No. 43755301), July 29, 1995, Unpublished.

SPONSOR: The Chlorpicrin Manufacturers Task Force c/o Nicklor Chemical Co., Long Beach CA

EXECUTIVE SUMMARY:

In an inhalation oncogenicity study (MRID 43755301), groups of 50 male and 50 female Sprague-Dawley CD rats were exposed (whole body) to chloropicrin vapor (99.6% pure) for up to 108 weeks at concentrations of 0, 0.1, 0.5, or 1.0 ppm. Toxicity was monitored by clinical observations, body weights and gains, food consumption, ophthalmology examinations, hematology, organ weights, necropsy observations and microscopic evaluation.

In the 0.5 and 1.0 ppm groups, there was a dose-dependent decrease in survival time and an increase in mortality rate in males. Survival at week 108 was 34% and 30% in males exposed at 0.5 or 1.0 ppm compared to 58% in controls and mean survival times were 672 and 647 days compared to 696 days for controls ($p<0.05$ and 0.01). Based on mortality and longevity, females appeared to be more tolerant than males in this study, an observation consistent with a previously submitted 90 day inhalation study.

Minimal but significant effects on body weights and gains were noted for both sexes but only in the first 2 weeks of the study. Males exposed at 1.0 ppm, had an increased incidence of rhinitis in the anterior nasal cavity (70% vs 40% in controls). There were no statistical increases in neoplasm incidence at any site and the incidence of neoplasms in all groups of males and females was similar to that normally seen in rats of this strain.

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However, one male and one female from the high dose groups were found to have a mast cell sarcoma of the larynx, which was cited by the reviewing pathologist as being a very rare tumor in rats. The significance of this tumor was considered uncertain.

The systemic LOEL is 0.5 ppm, based on decreased survival (males) and marginally decreased body weight gains in the first few weeks of the study (both sexes). The NOEL for males was 0.1 ppm and 1.0 ppm for females. Under the conditions of the study there was no oncogenic response. There were no increases in neoplasm incidence at any site and the incidence of neoplasms in all groups of males and females was similar to that normally seen in rats of this strain.

This chronic toxicity study in the rat is acceptable and does satisfy the guideline requirement for a chronic inhalation study (83-2a).

COMPLIANCE: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Chloropicrin
   Description Clear colorless oily liquid
   Lot/Batch #: 920130-2 (composite of samples from three manufacturers)
   Purity: 99.6 % a.i.
   Stability of compound: Validated as completely stable over the study.
   CAS #: 76-06-2

   Cl_3-C-NO_2

2. Vehicle and/or positive control: None

3. Test animals: Species: Rat
   Strain: Charles River CD (Sprague Dawley)
   Age and weight at study initiation: 6-5-7.5 weeks; mean group weights were 150--151 g in females and 206-207 g in males.
   Source: Charles River Laboratories, Portage MI
   Housing: Two/cage during the first 7 days of acclimation and singly thereafter.
   Diet: Purina Certified Rodent Chow #5002 (ground) ad libitum
   Water: Westmoreland Co. Municipal tap water ad libitum
   Environmental conditions: Temperature: 66-76°F

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Air changes: 14/hour during exposure
Photoperiod: 12 hour light/dark
Acclimation period: Three weeks

B. STUDY DESIGN:

1. In life dates: - start:8/10/92 end:9/1/94

2. Animal assignment:

Animals were assigned using a stratified randomization method based on body weights to the test groups in Table 1. Animals were examined by a veterinarian before randomization and those considered unacceptable based on clinical signs or body weight gains were rejected.

TABLE 1: STUDY DESIGN

Test Group	Target Exposure Conc. (ppm)	Analyzed Mean Conc. (ppb)	Main Study 107 weeks No. of rats	
			male	female
Control	0	0	50	50
Low (LCT)	0.1	100 ± 6.1	50	50
Mid (MCT)	0.5	499 ± 21.9	50	50
High (HCT)	1.0	995 ± 44.3	50	50

3. Dose selection rationale:

The rationale was based on a 90-day study in groups of 10 male and 10 female rats exposed to chloropicrin vapor at concentrations of 0.3, 1.0, or 3.0 ppm. The 3-ppm level caused death in 3 males, and decreased body weight/food consumption during weeks 1-2 in both sexes. Increases in red blood cell parameters in males, and increases in lung weight were seen. Hyperplasia/dysplasia of the nasal respiratory epithelia was observed at 1.0 and 3.0 ppm and in the lungs, bronchitis/bronchiolitis and accompanying hyperplasia of the epithelia as well as hyperplasia and fibrosis of the peribronchiolar muscle were observed. The histologic changes were mild in the 0.3-ppm groups. The 3 ppm exposure level was considered to be too high for the chronic inhalation study.

4. Exposure methodology:

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Inhalation chamber - Rectangular stainless steel and glass chambers with a volume of 4320 l were used. The airflow, monitored with a pressure gauge, was ca. 1000 l/min (14 changes/hour). The calculated time to reach 99% of target concentration was 20 minutes.

Vapor generation- Liquid chloropicrin was metered from a syringe into a heated glass evaporator with a temperature monitor. Vapor was diluted with filtered chamber air to achieve the desired concentration of test substance. The rate of airflow to the chambers was monitored continuously and recorded every 30 minutes to assure an oxygen content of at least 19%. During the first week of the study, a TSE Aerodynamic Particle Sizer was used to check for the possibility of an aerosol (highest level).

Chamber monitoring - For 3 days prior to the study, chamber chemical concentrations were monitored for consistency of vapor concentration. Uniformity of distribution was measured at pretest and at 6-month intervals by sampling at 5 positions in each chamber (3 concentrations) at 4 time periods. A gas chromatography-electron capture detection method was used for chemical analysis. During each exposure period, atmospheric analyses were conducted 9 times in the breathing zone of the rats. Daily nominal concentrations were also determined. Chamber temperature and humidity were constantly monitored and were recorded 12 times during each 6-hour exposure period. At 2 intervals during the study, chambers (1.0 ppm) were monitored to determine if phosgene (a thermal decomposition product of chloropicrin) was detectible.

Results:

The means of the daily mean chamber concentrations are summarized in Table 1. No detectible chloropicrin was measured in control chambers and no phosgene (detection limit 20 ppb) was seen at the highest concentration. All analyzed values were close to nominal. No aerosol was present and tests for uniformity of vapor concentrations indicated a coefficient of values of less than 3%

The means of daily mean temperature values ranged from 22.7-23.2°C and humidity values ranged from 47.1-49.0% at exposure concentrations of 0, 0.1, 0.5, and 1.0 ppm.

5. Statistics:

Quantitative data for continuous variables were tested for equality of variances using Levene's test and this

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was followed by ANOVA and t-tests. When data had similar variances, a pooled t-test was used for pairwise comparison. ANOVA for unequal variance was used for heterogeneous data. Nonparametric data were evaluated with the Kruskal-Wallis test followed by the Mann-Whitney U-test.

Incidence data were compared using the Fisher exact test. Mortality data and incidence and mean time to first palpable mass were analyzed by product limit survival analysis.

C. METHODS:

1. Observations:

Animals were inspected daily for signs of toxicity and 2x/day for mortality. Detailed individual animal examinations were conducted weekly. During the exposures, observations were recorded on a group basis but individual animal signs of toxicity observed preceding or after exposure were recorded for individual rats. Palpable masses were recorded as to time of onset and their progression or regression was followed.

2. Body weight:

Animals were weighed weekly for the first 13 weeks and every other week thereafter.

3. Food consumption:

Measured weekly for the first 13 weeks and every 5th week thereafter.

4. Ophthalmoscopic examination:

Eyes were examined prior to the first exposure and at 107 weeks with an indirect ophthalmoscope or a slit lamp following dilation with Mydriacil® (tropicamide 1%).

5. Blood was collected for hematology from all surviving animals (non-fasted) at 12, 18 and 24 months by retroorbital bleeding. The CHECKED (X) parameters were examined at 18 and 24 months. Blood smears for total and differential white cell counts were obtained from all animals at 12 months.

a. Hematology:

x	Hematocrit (HCT)*	x	Leukocyte differential count*
x	Hemoglobin (HGB)*	x	Mean corpuscular HGB (MCH)
x	Leukocyte count (WBC)*	x	Mean corpusc. HGB conc.(MCHC)
x	Erythrocyte count (RBC)*	x	Mean corpusc. volume (MCV)
x	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Required for chronic studies based on Subdivision F Guidelines

b. Clinical Chemistry: Not in protocol.

6. Sacrifice and Pathology:

All animals that died and those sacrificed on schedule were subjected to gross pathological examination and the CHECKED (X) tissues were collected for histological examination. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue	x	Aorta*	xx	Brain*
x	Salivary glands*	x	Heart*	x	Periph.nerve*
x	Esophagus*	x	Bone marrow*	x	Spinal cord (3 levels)*
x	Stomach*	x	Lymph nodes*		
x	Duodenum*	xx	Spleen*	x	Pituitary*
x	Jejunum*	x	Thymus*	x	Eyes (optic n.)*
x	Ileum*				
x	Cecum*		UROGENITAL		GLANDULAR
x	Colon*	xx	Kidneys*+	xx	Adrenal gland*
x	Rectum*	x	Urinary bladder*	x	Lacrimal gland
xx	Liver*+	xx	Testes*+	x	Mammary gland*
	Gall bladder*	x	Epididymides	x	Parathyroids*++
x	Pancreas*	x	Prostate	x	Thyroids*++
		x	Seminal vesicle		Zymbal gland
	RESPIRATORY	x	Ovaries*+		
x	Trachea*	x	Uterus*		OTHER
xx	Lung*	x	Vagina	x	Bone*
x	Nose			x	Skeletal muscle*
	Pharynx			x	Skin*
x	Larynx			x	All gross lesions and masses*

* Required for chronic studies based on Subdivision F Guidelines.
+ Organ weight required in chronic studies.
++ Organ weight required for non-rodent studies.

Microscopic examination was conducted on all the above tissues in the control and high exposure groups. The lungs,

trachea, larynx, nasal tissues, liver, kidneys and tissues with gross lesions were examined in all animals in the low and mid concentration groups. The mammary glands and ovaries were also examined from low and mid exposure level females.

II. RESULTS

A. OBSERVATIONS:

1. Toxicity - An increase in hypoactivity and a reduced startle reflex during exposure at 1.0 ppm were reported. During non-exposure periods, no clinical signs were considered related to exposure.

2. Mortality - Table 2 summarizes mortality and percent survival at selected study intervals.

Table 2. Mortality and (Percent Survival)[a]

Week	Exposure Concentration (ppm)							
	Males				Females			
	0	0.1	0.5	1.0	0	0.1	0.5	1.0
52	1 (98)	3 (94)	1 (98)	1 (98)	1 (98)	0(100)	0(100)	3(94)
78	4 (92)	7 (86)	5 (90)	9 (82)	6 (88)	8 (84)	9 (82)	7(86)
89	10(80)	12(76)	13(74)	15(70)	11(78)	15(70)	17(66)	16(68)
100	14(72)	21(58)	25(50)	30(40)	20(60)	25(50)	23(54)	24(52)
107	21(58)	29(42)	33(34)	35(30)	25(50)	32(36)	29(42)	28(49)
Mean Survival Time (days)								
	696	669	672*	647**	697	673	670	661

Source: Extracted from Study 92N1106 Appendix 6 (pp 937-946) and Table 1,p25.
* Significantly lower than in control group, $p<0.05$.
**Significantly lower than in control group $p<0.01$.
a Percent (calculated by reviewer) is based on 50 rats/sex/ group and not corrected for 2 accidental deaths, one control female and one female at 0.5 ppm.

The mortality rates in male rats were increased in a concentration dependent manner and were 42, 58, 66, and 70% in the 0, 0.1, 0.5, and 1.0 ppm exposure groups. Mean survival time was significantly lowered in males at 0.5 and 1.0 ppm. No significant effect on either parameter was seen in exposed females.

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B. BODY WEIGHT:

No exposure related effects on mean absolute body weights were observed in either males or females. Mean body weights in males exposed at 1.0 ppm were 4 and 3% lower than controls at weeks 1 and 2 and 2% lower at both weeks in females exposed at 1.0 ppm. Table 3 summarizes data on body weight gains. Weight gains at weeks 1 and 2 were 28 and 8% lower in males at 1.0 ppm than in controls ($p < 0.01$) and 8% decreased at week 1 in the 0.5-ppm group. Body weight gains (cumulative) in 1.0-ppm females were 25, 12.4 and 7% lower than in controls at weeks 1, 2, and 7 ($p < 0.01$); in the 0.5-ppm group of females gains were 15, 9.5, and 8% lower ($p < 0.01$). Cumulative weight gains tended to be 3-7% higher than in controls from weeks 17-53 in the male group exposed at 0.5 ppm.

Table 3. Mean Body Weight Gains in Rats Exposed to Chloropicrin

Weeks	Exposure Concentration (ppm)							
	Males				Females			
	0	0.1	0.5	1.0	0	0.1	0.5	1.0
0-1	31.2	30.1	28.6*	22.6*	15.8	14.1	13.4*	11.9*
0-2	78.0	77.3	77.7	71.4*	37.1	34.8	33.6*	32.5*
0-7	214	216	220	211	96.8	90.8†	89.5*	89.9*
0-13	288	292	296	285	127	122	119	123
0-101	458	436	489	478	354	295	315	344

Source: Study 92N1106, Tables 6 and 8, pp 58 and 68.
* Significantly different from control value, $p < 0.01$; † p,0.05.

C. FOOD CONSUMPTION:

No exposure related effects on food consumption were observed.

D. OPHTHALMOSCOPIC EXAMINATION:

No abnormal findings were observed in the exposed groups.

E. BLOOD WORK:

1. Hematology: No exposure related effects were observed. All parameters were within the normal range.

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2. Clinical chemistry: Not required by protocol.

F. URINALYSIS: Not conducted.

G. SACRIFICE AND PATHOLOGY:

1. Organ weight: No statistically significant effects on absolute or relative organ weights were observed in males. In females no significant effects on absolute organ weights were observed; slight but significant ($p<0.05$) decreases for organ-to-body weight ratios of liver and kidneys at 0.5 ppm were the result of an 11% lower (nonsignificant) mean terminal body weight. In males, but not females at 1.0 ppm, a nonsignificant increase in absolute lung weight (17% higher than control) and lung-to- body weight ratio (10%) was observed. This may be related to exposure but its importance is equivocal.

2. Gross pathology: No gross lesions at necropsy could be attributed to exposure. Hyperinflation of the lungs was observed in 4, 12, 10, and 12% of males and 6, 8, 6, and 4% of females exposed at 0, 0.1, 0.5, or 1.0 ppm, respectively. Swollen livers were observed in 42-58% of males in various groups and 32-44% of females in various groups, but no exposure-related trend was present. Pituitary masses and associated brain depression were frequent in male groups (34-46%) and female groups (66-80%) and mammary masses were frequent in female groups (34-50%) but no exposure trend was apparent.

3. Microscopic pathology:

a) Non-neoplastic - The only exposure related nonneoplastic lesion observed microscopically was rhinitis in the anterior nasal cavities of male rats exposed at 1.0 ppm. Table 4 shows the incidence of the finding for animals found dead/sacrificed moribund, for those sacrificed at 107-108 weeks and for all animals. The lesion was characterized by sporadic lymphocytic or neutrophilic mucosal/submucosal infiltrates and occasionally by purulent exudate. The incidence was significantly increased ($p<0.01$) for all 1.0 ppm males (70%) compared to controls (40%) and for 1.0-ppm males at terminal sacrifice the incidence was 87% ($p<0.05$) compared to 55% for controls. The severity of rhinitis

Table 4. Incidence of Rhinitis in Nasal Cavity of Rats

Sex/Fate	Exposure Concentration (ppm)			
	0	0.1	0.5	1.0
Males				
Dead/moribund	4/21	14/29	13/33	22/35
Terminal Sacr.	16/29	10/21	8/17	13/15*
All	20/50	24/50	21/50	35/50**
Females				
Dead/moribund	9/25	11/32	15/29	12/28
Terminal Sacr.	9/25	6/18	11/21	11/22
All	18/50	17/50	26/50	23/50

Source: Study 92N1106 Tables 29-33, pp 161, 218, 288, 336, 376, and 425.
* Significantly different from control $p<0.05$; ** $p<0.01$.

was somewhat increased in 1.0-ppm males (moderate to marked in 40% of cases at 1.0 ppm compared to 30% of cases in controls). No increased incidence of significance was observed in exposed females (Table 4). Submucosal mineralization/ossification and epithelial hyaline inclusions in the nasal tissue (predominantly minimal/mild in grade) were fairly frequent in all groups of males and females but there was no clear exposure related trend in either sex. The incidence of minimal arterial mineralization in the lungs was significantly increased in males exposed to 0.1 ppm or 0.5 ppm (62 and 66%), but was only 14% in controls and 30% in high exposure males; the incidence in all female groups ranged from 50-60%. This finding in males is not considered related to exposure or of importance. No exposure related changes were observed at other organ sites.

b) Neoplastic - No exposure related increases in any neoplasms were seen in the study. The most frequently occurring neoplasm was pituitary adenoma which was present at an incidence of 56-72% in male groups and 76-92% in female groups. For all females in the study, the incidence of mammary fibroadenoma was 20%,32%,28% and 32% at exposure levels of 0, 0.1, 0.5, or 1.0 ppm and the incidence of mammary adenocarcinoma was 32%, 24% ,18%, and 12% at the same exposure levels. The number of females with adenoma/fibroadenoma/adenocarcinoma of the mammary gland (derived by the reviewer from the individual animal pathology sheets was 23, 24, 23,and 21

in the control, 0.1-ppm, 0.5-ppm or 1.0-ppm groups. It was concluded that the number of mammary tumor bearing females was similar in all groups.

III. DISCUSSION

A. Study Authors Conclusions - The study authors concluded that exposure of rats to chloropicrin for 24 months resulted in few clinical signs of toxicity and these signs were only in the group exposed to 1.0 ppm. Minimal effects on body weight or body weight gain occurred at 0.5 and 1.0 ppm and these effects were only noted in the first 2 weeks of the study. The only nonneoplastic lesion noted microscopically was rhinitis in the nasal cavity of the male rats exposed at 1.0 ppm. No increases in neoplastic lesion incidence were considered exposure related. While there were no group differences in the relative frequency of cause of death, mean survival time was decreased in males in the 0.5 and 1.0 ppm groups. The NOEL for systemic effects was 0.1 ppm.

Reviewers' Discussion - The rationale for dose selection was defensible. The minimal effects on body weights and weight gains in the first 2 weeks of the study may have been associated with adaption of the animals to exposure, however, according to the protocol animals were acclimated to the exposure chambers for 2 days in the week prior to study initiation.

The survival rate of control males in the present study (60% at 104 weeks and 58% at 108 weeks) is relatively high for this strain of rat. The study authors reported that the mortality rates in the 0.1 and 0.5 ppm groups of males in the present study were within the historical range for controls in previous chronic studies in the testing laboratory. The BBRC laboratory historical data were not submitted but the mean survival rate in untreated rats of the same strain and source in 15 2-year studies was 44% (Charles River Laboratories, 1994).

The increased mortality rate in males in 0.5 and 1.0 ppm (not seen in females) has a high level of significance and there is also a clear exposure related trend. The females rats in this study may have been able to tolerate a slightly higher exposure level. In a previously submitted 90 day inhalation study in rats (MRID# 43063201), mortality in males and females were comparable (0/10 for each sex) in the controls, 0.3 and 1.0 ppm test groups and 0/10 for the females of the 3.0 ppm (HD) group. Three of 10 males died in the 3.0 ppm test group of causes which could be related to the administration of CP. One of 10 females in the

control group died of lymphosarcoma, unrelated to CP administration.

The histologic changes in the nasal cavity of males exposed at 1.0 ppm are a portal of entry effect and not a systemic effect. No toxicologically important effects on the lungs were observed in this study. It was noted that arterial mineralization of the lungs (all grades) was significantly increased ($p<0.01$) in males exposed at 0.1 and 0.5 ppm; however, the severity was minimum in 31/36 and 33/37 of these males and incidence of mild/moderate/marked mineralization was 10, 10, 8 and 6% in males in the 0, 0.1, 0.5, or 1.0-ppm groups. Incidence in all groups of females was 50-60%. We do not consider the finding related to dosing. Although the study authors reported hypoactivity and a reduced startle activity in 1.0 ppm animals during exposure, we could not verify the frequency of the occurrence.

\cprtonc.der E1 October 7, 1996

DATA EVALUATION REPORT

CHLOROPICRIN

Study Type: 83-2b; Inhalation Oncogenicity Study in Mice

Dynamac Study No. 123B (MRID 436322201)

Prepared for

Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
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Prepared by

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Disclaimer

This Data Evaluation Report may have been altered by the Health Effects Division subsequent to signing by Dynamac Corporation personnel.

EPA Reviewer: Stanley Gross, Ph.D. Stanley Gross, Date 10/17/96
Review Section II, Toxicology Branch I (7509C)
EPA Secondary Reviewer: Marion Copley, D.V.M. Marion Copley, Date 11/13/96
Review Section IV Toxicology Branch I (7509C)

DATA EVALUATION RECORD

STUDY TYPE: Inhalation Oncogenicity Study-Mouse;OPPTS 870.4100 §83-2b
DP BARCODE: D215526 SUBMISSION CODE:S487231
P.C. CODE: 081501 TOX. CHEM. NO.:214
TEST MATERIAL (PURITY): Chloropicrin (99.6%)

SYNONYMS: Nitrochloroform, trichloronitromethane

CITATION: Burleigh-Flayer, W.J. Kintigh, and C.L. Benson (1995) Chloropicrin: Vapor Inhalation Oncogenicity Study in CD-1® Mice (MRID No.43632201); Bushy Run Research Center, Export PA 15632. Laboratory project No. 92N1105; April 25, 1995; Unpublished.

Robert M. Kovatch, 1996). Pathology Peer REview of Chloropicrin: Vapor Inhalation Oncogenicity Study in CD-1(R) Mice. Study No. 92N1105. Pathology Associates International Corporation, Frederick, MD. January 31, 1996. Unpublished.

SPONSOR: The Chloropicrin Manufacturers Task Force c/o Nicklor Chemical Co., Long Beach CA

EXECUTIVE SUMMARY:

In a vapor inhalation oncogenicity study (MRID 43632201), groups of CD-1® mice (50 mice/sex) were exposed to concentrations of 0 (air), 0.10, 0.50 or 1.0 ppm chloropicrin vapor (99.6% pure) for 6 hours/day, 5 consecutive days/week for 78 weeks. Toxicity was monitored by clinical observations, body weights and gains, food consumption, ophthalmology examinations, hematology, organ weights, necropsy observations and microscopic evaluation.

There were no toxic signs or mortality associated with the CP exposures. In males and females exposed to 0.5 ppm, mean body weights were lower and cumulative weight gains were depressed throughout the study. At the 1-ppm exposure level, mean body weights/weight gains at week 27 were significantly ($p<0.01$) decreased (5%/19% in males and 9%/32% in females). Food consumption was decreased in females. At week 27, mean weights were decreased 5% and 9% as compared to controls in males and females,respectively, and weight gains were 13% and 24% lower. Food consumption in females was 5% decreased (weeks 1-13). At sacrifice, lung weights were increased 12% (males) and 17%

(females). In the nasal cavity, rhinitis with serous exudate was significantly increased in both sexes and hyaline epithelial inclusions and olfactory epithelial atrophy were significantly increased in females.

Non-neoplastic lesions related to dosing with CP related to the lungs and the nasal cavity. Lung bronchiectasis characterized by dilation of the bronchus was accompanied by thickened bronchial wall (fibrosis) and minimal inflammatory exudates elevated in both sexes exposed to 0.5 and 1.0 ppm. In the nasal cavity, serous exudate and atrophy of the olfactory epithelium, decreased olfactory neurons were increased in both sexes exposed to 0.5 and 1.0 ppm. The lesions of the nasal cavity seen at 0.5 ppm were increased in incidence in both sexes and olfactory epithelial atrophy was present in 72-80% of the mice. In addition to the lung lesions at 0.5 ppm, alveolar histiocytosis was observed in 29/50 males and 35/50 females ($p<0.01$) and alveolar/ bronchiolar cell hyperplasia was present in 6/50 1-ppm females ($p<0.05$).

Neoplastic findings -- incidence of pulmonary edema were slightly increased but not significant statistically, in male and female mice of the 0.5 and 1.0 ppm groups. The slightly elevated indices in 0.5 and 1.0 ppm male and female groups were attributed to random variation.

The LOEL for systemic toxicity and irritation is 0.5 ppm based on lower body weight and body weight gains, increases in lung weight and histologic changes in the nasal cavity, upper respiratory tract and lungs. The systemic NOEL is 0.1 ppm. CP was not neoplastic in this study.

This oncogenicity study in the mouse is acceptable and satisfies the guideline requirement for an oncogenicity study in mice (83-2b).

COMPLIANCE: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

I. MATERIALS AND METHODS

A. MATERIALS:

1. Test Material: Chloropicrin
   Description Clear colorless oily liquid
   Lot/Batch #: 920130-2 (composite of samples from three manufacturers)
   Purity: 99.6 % a.i.
   Stability of compound: Validated as completely stable

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CAS #: 76-06-2

Structure: Cl_3-C-NO_2

2. Vehicle and/or positive control: None

3. Test animals: Species: Mouse
Strain: Charles River CD-1
Age and weight at study initiation: approximately 6.6-7 weeks old, males 27-38g, females 20-29g.
Source: Charles River Laboratories, St. Constant, Quebec, Canada
Housing: Two/cage during the first 7 days of acclimation and singly thereafter.
Diet: Purina Certified Rodent Chow #5002 (ground) ad libitum
Water: Westmoreland Co. Municipal tap water ad libitum
Environmental conditions: Temperature: 66-77°F
Humidity: 40-70%
Air changes: 14/hour during exposure; ≥ 10/hr. in animal rooms
Photoperiod:12 hour light/dark
Acclimation period: approximately 3 weeks

B. STUDY DESIGN:

1. In life dates: start exposures: August 10, 1992, sacrifice: males 2/28/94-3/2/94, females 3/2/94-3/3/94

2. Animal assignment:

Animals were assigned using a stratified randomization method based on body weights to the test groups in Table 1. Animals were examined by a veterinarian before randomization and those considered unacceptable based on clinical signs or body weight gains were rejected.

3. Dose selection rationale:

The rationale was based on a 90-day study in groups of 10 male and 10 female mice exposed to chloropicrin vapor at concentrations of 0.3,1.0, or 3.0 ppm. The 3-ppm level caused death in 3/10 males. Decreased body weight/body weight gain were seen throughout the study in both sexes of mice at 1.0 and 3.0 ppm and this was accompanied by decreased food consumption. Increases in red blood cell parameters in males, and increases in lung weight were seen. Hyperplasia/dysplasia of the nasal respiratory epithelia was observed at 1.0 and 3.0 ppm and in the lungs, bronchitis/bronchiolitis and accompanying

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hyperplasia of the epithelia as well as hyperplasia and fibrosis of the peribronchiolar muscle were observed. Two 0.3-ppm females had mild hyaline inclusions in nasal tissue. The 3-ppm exposure level was considered to be too high for the chronic inhalation study.

TABLE 1: STUDY DESIGN

Test Group	Target Exposure Conc. (ppm)	Analyzed Mean Conc. (ppb)	Main Study No. of rats	
			male	female
Control	0	0	50	50
Low (LCT)	0.1	100 ± 6.4	50	50
Mid (MCT)	0.5	498 ± 22.3	50	50
High (HCT)	1.0	988 ± 43.4	50	50

4. Exposure methodology:

Inhalation chamber - Rectangular stainless steel and glass chambers with a volume of 4320 l were used. The airflow, monitored with a pressure gauge, was ca 1000 l/min (14 changes/hour). The calculated time to reach 99% of target concentration was 20 minutes.

Vapor generation- Liquid chloropicrin was metered from a syringe into a heated glass evaporator with a temperature monitor. Vapor was diluted with filtered chamber air to achieve the desired concentration of test substance. The rate of airflow to the chambers was monitored continously and recorded every 30 minutes to assure an oxygen content of at least 19%. During the first week of the study, a TSE Aerodynamic Particle Sizer was used to check for the possibility of an aerosol (highest level).

Chamber monitoring - For 3 days prior to the study, chamber chemical concentrations were monitored for consistency of vapor concentration. Uniformity of distribution was measured at pretest and at 6-month intervals by sampling at 5 positions in each chamber (3 concentrations) at 4 time periods. A gas chromatography-electron capture detection method was used for chemical analysis. During each exposure period, atmospheric analyses were conducted 9 times in the breathing zone of the rats. Daily nominal concentrations were also

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determined. Chamber temperature and humidity were constantly monitored and were recorded 12 times during each exposure. At 52 weeks, chambers (1.0 ppm) were monitored to determine if phosgene (a thermal decomposition product of chloropicrin) was detectible. A commercial phosgene detection tube with a sensitivity of 0.02 ppm was used.

Results: The means of the daily mean chamber concentrations are summarized in Table 1. No detectible chloropicrin was measured in control chambers and no phosgene (detection limit 20 ppb) was seen at the highest concentration. All analyzed values were close to nominal. No aerosol was present and tests for uniformity of vapor concentrations indicated coefficients of variance of less than 2.3%

The means of daily mean temperature values ranged from 22.9-23.5°C and humidity values ranged from 47.5-49.1% at exposure concentrations of 0, 0.1, 0.5, or 1.0 ppm.

5. Statistics:

Quantitative data for continuous variables were tested for equality of variances using Levine's test and this was followed by ANOVA and t-tests. When data had similar variances, a pooled t-test was used for pairwise comparison. ANOVA for unequal variance was used for heterogeneous data. Nonparametric data were evaluated with the Kruskal-Wallis test followed by the Mann-Whitney U-test.

Incidence data were compared using the Fisher exact test. Mortality data and incidence and mean time to first palpable mass were analyzed by product limit survival analysis.

C. METHODS:

1. Observations: Animals were inspected daily for signs of toxicity and 2x/day for mortality. Detailed individual animal examinations were conducted weekly. During the exposures, observations were recorded on a group basis but individual animal signs of toxicity observed preceding or after exposure were recorded for individual rats. Palpable masses were recorded as to time of onset and their progression or regression was followed.

2. Body weight: Animals were weighed weekly for the first 13 weeks and every other week thereafter.

3. Food consumption: Measured weekly for the first 13 weeks and every 5th week thereafter.

4. Ophthalmoscopic examination: Not required.

5. Blood was collected for hematology from all surviving animals (non-fasted) at 18 months by retroorbital bleeding. The CHECKED (X) parameters were examined. Blood smears were obtained from all animals at 12 months and total and differential white cell counts evaluated for the control and high-dose groups.

a. Hematology:

x	Hematocrit (HCT)*	x	Leukocyte differential count*
x	Hemoglobin (HGB)*	x	Mean corpuscular HGB (MCH)
x	Leukocyte count (WBC)*	x	Mean corpusc. HGB conc.(MCHC)
x	Erythrocyte count (RBC)*	x	Mean corpusc. volume (MCV)
x	Platelet count*		Reticulocyte count
	Blood clotting measurements*		
	(Thromboplastin time)		
	(Thromboplastin time)		
	(Clotting time)		
	(Prothrombin time)		

* Required for chronic studies based on Subdivision F Guidelines

b. Clinical Chemistry: Not in protocol.

7. Sacrifice and Pathology:

All animals that died and those sacrificed on schedule were subjected to gross pathological examination and the CHECKED (X) tissues were collected for histological examination. The (XX) organs, in addition, were weighed.

	DIGESTIVE SYSTEM		CARDIOVASC./HEMAT.		NEUROLOGIC
	Tongue	x	Aorta*	xx	Brain*
x	Salivary glands*	x	Heart*	x	Periph.nerve*
x	Esophagus*	x	Bone marrow*	x	Spinal cord (3 levels)*
x	Stomach*	x	Lymph nodes*		
x	Duodenum*	xx	Spleen*	x	Pituitary*
x	Jejunum*	x	Thymus*	x	Eyes (optic n.)*
x	Ileum*				
x	Cecum*		UROGENITAL		GLANDULAR
x	Colon*	xx	Kidneys*+		
x	Rectum*	x	Urinary bladder*	xx	Adrenal gland*
xx	Liver*+	xx	Testes*+	x	Lacrimal gland
x	Gall bladder*	x	Epididymides	x	Mammary gland*
x	Pancreas*	x	Prostate	x	Parathyroids*++
		x	Seminal vesicle	x	Thyroids*++
	RESPIRATORY	x	Ovaries*+		
x	Trachea*	x	Uterus*		OTHER
xx	Lung*	x	Vagina	x	Bone*
x	Nose			x	Skeletal muscle*
	Pharynx			x	Skin*
x	Larynx			x	All gross lesions and masses*

* Required for chronic studies based on Subdivision F Guidelines.

Microscopic examination was conducted on all the above tissues in the control and high exposure groups. The lungs, trachea, larynx, nasal turbinates, liver, kidneys and tissues with gross lesions were examined for all animals in the low- and mid-concentration groups.

II. RESULTS

A. OBSERVATIONS:

1. Toxicity: During exposures, one mouse in the 1.0-ppm exposure group exhibited stereotypy, hyperactivity, circling, hunched posture and clonic convulsions. A second mouse in the 0.10 ppm exposure group exhibited stereotypy, hyperactivity, and circling. Since both these mice exhibited these signs only during one exposure session, they were not considered to be of toxicological significance. During nonexposure periods, male mice in the exposed groups exhibited a slightly higher incidence of swollen periocular tissue than the control group mice.

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This finding was not considered to be of toxicological significance. No other treatment-related signs of toxicity were reported in this study.

2. Mortality: Table 2 summarizes mortality and percent survival at selected study intervals. No significant differences were seen and at 18 months; survival in all groups was within the normal range expected for this strain of mouse.

B. BODY WEIGHT:

Both sexes of mice in the 0.50 and 1.0 ppm exposure groups showed concentration-related decreases in mean body weights and body weight gains throughout the study. Mean body weights during the first 53 weeks were 3-6% decreased in 1.0-ppm males and 4-10% decreased in 1.0-ppm females when compared to controls. At 0.5 ppm, the mean body weights at week 53 were 3% and 4% decreased in males and females, respectively. Body weight gains at week 13 were 15% and 23% lower than control in 0.5-ppm and 1.0-ppm males and 12% and 21% decreased in females exposed at 0.5 and 1.0 ppm, respectively.

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TABLE 2. Mortality and (Percent Survival)

Week	Exposure Concentration (ppm)							
	Males				Females			
	0	0.1	0.5	1.0	0	0.1	0.5	1.0
53	1 (94)	2 (90)	5 (90)	0 (100)	3 (92)	3 (92)	3 (98)	4 (88)
67	8 (84)	8 (84)	9 (81)	7 (90)	9 (81)	10 (84)	7 (91)	9 (85)
81	14(72)	22(56)	18 (64)	8 (86)	11(72)	12 (70)	16(68)	12(68)
Mean Survival Time (days)								
	525	516	515	552	530	526	550	523

Data were extracted from BBRC Report No. 92N1105 Table 1 (p 25) and Appendix pp791-9.
a. In calculating percent survival, the animals with accidental deaths were excluded: 0, 1, 2, and 2 in male groups and 2, 1, 3, and 2 in female groups.

TABLE 3. Mean Body Weight Data in Mice Exposed to Chloropicrin

	Males				Females			
	0	0.1	0.5	1.0	0	0.1	0.5	1.0
	Mean body weight (g)							
Weeks 0	31.9	31.8	31.6	31.8	23.9	24.1	24.3	24.2
6	37.3	37.6	36.4	36.2*	28.4	28.8	27.8	27.3*
13	40.3	39.4	38.8*	38.4**	30.6	31.0	30.2	29.5**
27	42.6	41.5	41.0*	40.5**	33.2	32.5	31.3**	30.3**
53	43.3	43.1	41.9	40.5**	34.9	34.7	33.5*	31.4**
81	41.9	43.1	41.7	40.3	36.1	35.8	34.0**	32.5**
Cumulative body weight gain (g)								
0-6	5.5	5.9	4.8*	4.4*	4.5	4.7	3.5**	3.1**
0-13	8.5	7.7	7.2**	6.6**	6.7	6.3	5.9*	5.3**
0-27	10.7	9.8	9.3*	8.7**	9.3	8.8	7.1*	6.4**
0-53	11.3	11.2	10.4	8.6	10.9	10.5	9.3**	7.1**
0-81	10.2	11.3	9.9	8.8	12,2	11.6	9.7**	8.3**

Data extracted from BBRC Report No. 92N1105, Tables 4-7 (pp 50-66).
* Significantly different from control value, $p<0.05$; ** $p<0.01$

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C. FOOD CONSUMPTION:

There were no compound related effects on food consumption in exposed males; In the first 13 weeks consumption in 1-ppm males was 2% lower than in controls. Food consumption in females exposed at 0.5 and 1 ppm was decreased during many weeks of the study. Over the first 13 weeks, consumption was 5 and 10% lower than in controls at 0.5 and 1.0 ppm, respectively.

D. BLOOD WORK:

1. Hematology: No exposure related effects were observed. All parameters were within the normal range.

2. Clinical chemistry: Not required by protocol.

E. SACRIFICE AND PATHOLOGY:

1. Organ weights: Mean weights of lungs were significantly increased compared to controls in males at 0.5 ppm (14%) and 1 ppm (16%) and in females exposed at 1 ppm (36%). Lung-to-body weight ratios were significantly increased in both sexes exposed at 0.5 and 1 ppm (Table 4). No other organ weight changes were observed in the study.

TABLE 4. Absolute and Relative Lung Weghts in Mice Exposed to Chloropicrin for 18 Months

Weight (g/%)	Exposure Concentration (ppm)			
	0	0.1	0.5	1
	Males			
Absolute (g)	0.33	0.32	0.37*	0.38**
Relative (%)	0.81	0.76	0.90	0.94**
	Females			
Absolute (g)	0.30	0.35	0.35	0.40**
Relative (%)	0.83	1.00	1.03*	1.26**

Data extracted from BBRC Report No. 92N1105, Tables 14 and 15 (pp 78-79).

* Significantly different from control value, $p<0.05$; ** $p<0.01$.

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2. Gross pathology: There were no treatment-related gross pathological effects seen in this study.

3. Microscopic pathology:

a) Non-neoplastic: Nonneoplastic lesions in the chloropicrin-treated mice in the 0.5 and 1 ppm exposure groups included serous exudate and minimum/moderate rhinitis of the nasal cavity, olfactory epithelial atrophy and hyaline epithelial inclusions in the cytoplasm of the respiratory and/or olfactory epithelium of the nasal cavities. In the respiratory tract, peribronchial lymphocytic infiltrates, bronchial submucosal fibrosis, and bronchiectasis (predominantly minimal/mild) were significantly increased in both sexes exposed at 0.5 and 1 ppm. Alveolar histiocytosis was significantly increased at 1 ppm (both sexes) and in 1-ppm females, alveolar protein deposits and bronchioalveolar cell hyperplasia were observed (Table 5). Lung lesions at termination were slightly more severe in females than in males. All these lesions were considered to result from chemical irritation from chloropicrin exposure and consequential responses to that irritation.

A small number of nonneoplastic lesions were seen at other sites. Adenitis of the auditory sebaceous glands was increased in 1-ppm males (not significant) as was corneal mineralization in 1-ppm females. Plasmacytosis of the submandibular lymph nodes, lymphoid hyperplasia of the spleen, and liver Ito-cell hyperplasia, were slightly increased in incidence of 1-ppm in females; however, these findings were not considered exposure related. Except for the chloropicrin exposure-related lesions in the nasal cavity and the lungs, the nonneoplastic lesions seen in this study were considered to be typical for this strain of mouse. A possible exception is the increased incidence of corneal mineralization in high exposure females (but not males). The incidence was 4/50, 2/50, 6/50 and 11/50 at 0, 0.1, 0.5 or 1.0 ppm.

TABLE 5. Nonneoplastic Lesions in the Nasal Tissue and Lungs of Mice Exposed to Chloropicrin[a]

Organ/Lesion	Exposure Concentration (ppm)							
	Males				Females			
	0	.0.1	0.5	1	0	0.1	0.5	1
Nasal Cavity	(50)	(50)	(49)	(50)	(50)	(50)	(50)	(50)
Serous exudate	4	7	18**	28**	4	5	36**	46**
Hyaline epithelial inclusions	3	6	7	16**	10	11	24**	37**
Rhinitis	6	7	17**	35**	3	16	18**	32**
Olfactory epithel. atrophy	5	6	8	40**	13	14	39**	36**
Lungs	(50)	(50)	(50)	(50)	(50)	(50)	(50)	(50)
Alveolar histiocytosis	18	17	22	29**	14	14	19	35**
Hemorrhage	4	4	10	12	8	10	8	13
Peribrochiolar lymph. infiltrat.	1	6	10**	12**	5	10	17	28
Bronchiectasis	0	3	28**	41**	0	5	28**	44**
Bronchial submuc. fibrosis	0	0	16**	19**	0	0	13**	22**
A/B cell hyperplasia	2	0	5	2	0	1	2	6*

a Data extracted from BBRC Report No. 92N1105, Table 28 (pp 216-221) and Table 32 (pp 359-365); the values in parentheses are the number of tissues examined.
* Significantly different from control incidence $p<0.05$; ** $p<0.01$.

b) Neoplastic: A slight but nonsignificant increase in the incidence of lung adenomas was seen in females exposed to 0.5 and 1.0 ppm. Table 6 shows the incidence of lung adenomas for all animals on study and animals at terminal sacrifice. One 0.5-ppm male and one 0.1-ppm female had both adenoma and carcinoma of the lung and one 1-ppm male had a malignant carcinosacoma in the lung. The incidence of animals with lung adenoma/carcinoma was 34, 28, 44 and 42% in males groups exposed at 0, 0.1, 0.5 and 1.0 ppm chloropicrin and 26, 26, 40, and 44% in females in the same groups, respectively. The increase in frequency of lung neoplasms in the 0.5 and 1.0 ppm groups was not considered to related to exposure. the incidence of hepatocellular adenoma in males was 12/50, 4/50 6/50 and 12/50. Neoplasms at other sites were incidental.

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TABLE 6. Neoplastic Lesions in the Lungs of Mice Exposed to Chloropicrin

Neoplasm	Exposure Concentration (ppm)							
	Males				Females			
	0	0.1	0.5	1.0	0	0.1	0.5	1.0
Adenoma All Terminal sac.	16/50 15/36	14/50 8/27	18/50 13/30	18/50 17/41	13/50 11/34	9/50 8/34	17/50 16/36	19/50 16/32
Carcinoma-All	1/50	0/50	5/50	2/50	0/50	4/50	3/50	4/50
Carcinosarcoma	0	0	0	1/50	1/50	2/50	2/50	3/50
Adenoma/ Carcinoma*	17/50	14/50	22/50	21/50	13/50	12/50	20/50	22/50

a Data extracted from BBRC Report No. 92N1105, Tables 29 and 33 (pp 226 and 370).

* This was determined by the reviewer from the individual animal data assuring that there was no double counting and represents the number of mice with one or both tumors.

III. **DISCUSSION**:

A. Study Author's Conclusions: Inhalation exposure to chloropicrin produced no significant effect on the incidence of neoplasms or on survival. Minimum to moderate non neoplastic lesions in the nasal cavity and lungs due to chemical irritation were seen at 0.5 and 1.0 ppm in both sexes (significant) and corneal mineralization of the eyes was observed in 1-ppm females.

B. PAIC Peer Review: The findings of the Pathology Associates International Corporation peer review were similar to that provided by the Author's report.

C. Reviewer's Discussion: The study was adequately conducted and survival was acceptable. The exposure levels appeared to be adequate to assess carcinogenicity. The concurrent control incidence of alveolar/bronchiolar adenoma in both sexes appears to be somewhat higher than normally observed for Charles River CD-1 mice. It would be useful if the sponsor provides the recent laboratory incidence of lung neoplasms in mice. We assess that there was no oncogenic response in this study in mice. The majority of alveolar/bronchiolar tumors that were observed were late developing neoplasms and not the cause of death. The

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NOEL for nonneoplastic effects in this is study is 0.1 ppm based on histologic lesions in the lungs and nasal tissues and on decreased weight gain in both sexes of mice.

cpmsonc.d2 E2 October 7, 1996