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BIOASSAY OF 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE FOR POSSIBLE CARCINOGENICITY

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U.S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
Public Health Service
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Carcinogenesis Testing Program
Division of Cancer Cause and Prevention
National Cancer Institute
National Institutes of Health
Bethesda, Maryland 20014

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FOREWORD: This report presents the results of the bioassay of 3-(chloromethyl)pyridine hydrochloride conducted for the Carcinogenesis Testing Program, Division of Cancer Cause and Prevention, National Cancer Institute (NCI), National Institutes of Health, Bethesda, Maryland. This is one of a series of experiments designed to determine whether selected environmental chemicals have the capacity to produce cancer in animals. Negative results, in which the test animals do not have a greater incidence of cancer than control animals, do not necessarily mean that the test chemical is not a carcinogen, inasmuch as the experiments are conducted under a limited set of circumstances. Positive results demonstrate that the test chemical is carcinogenic for animals under the conditions of the test and indicate that exposure to the chemical is a potential risk to man. The actual determination of the risk to man from animal carcinogens requires a wider analysis.

CONTRIBUTORS: The bioassay of 3-(chloromethyl)pyridine hydrochloride was conducted by Litton Bionetics, Inc., Kensington, Maryland, initially under direct contract to NCI and currently under a subcontract to Tracor Jitco, Inc., the prime contractor for the NCI Carcinogenesis Testing Program.

The experimental design and doses were chosen by Drs. E. K. Weisburger[1], J. H. Weisburger[1,2], and N. P. Page[1,3], NCI project officers, and Dr. F. M. Garner[4], the principal investigator. The administration of the test chemical and the observation of the animals were supervised by Dr. Garner and Mr. S. Johnson, the co-principal investigator, with the technical assistance of Mr. R. Cypher[4], Mr. H. D. Thornett[4], and Mr. D. J. Howard[4]. Ms. J. Blalock[4] was responsible for assembly of data.

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Animal pathology tables and survival tables were compiled at EG&G Mason Research Institute[5]. The statistical analyses were performed by Dr. J. R. Joiner[6] and Ms. P. L. Yong[6], using methods selected for the bioassay program by Dr. J. J. Gart[7]. Chemicals used in this bioassay were analyzed under the direction of Dr. E. Murrill[8], dosage solutions were analyzed by Mr. H. Paulin[4], and the results of the analyses were reviewed by Dr. S. S. Olin[6].

This report was prepared at Tracor Jitco[6] under the direction of NCI. Those responsible for the report at Tracor Jitco were Dr. L. A. Campbell, Director of the Bioassay Program; Dr. S. S. Olin, Deputy Director for Science; Dr. J. F. Robens, toxicologist; Dr. R. L. Schueler, pathologist; Dr. G. L. Miller, Ms. L. A. Waitz, and Mr. W. D. Reichardt, bioscience writers; and Dr. E. W. Gunberg, technical editor, assisted by Ms. Y. E. Presley and Ms. P. J. Graboske.

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SUMMARY

A bioassay of 3-(chloromethyl)pyridine hydrochloride for possible carcinogenicity was conducted by administering the test chemical by gavage to Fischer 344 rats and B6C3F1 mice.

Groups of 50 rats and 50 mice of each sex were administered 3-(chloromethyl)pyridine hydrochloride in a vehicle of distilled water three times per week at one of the following doses, either 75 or 150 mg/kg body weight for the rats and either 100 or 200 mg/kg body weight for the mice. The low-dose rats were dosed for 103 weeks and the low-dose mice for 102 weeks. Because of early deaths in the high-dose animals, the high-dose rats were dosed for only 83 weeks and the high-dose mice for only 81 weeks. Controls consisted of groups of 20 rats and 20 mice of each sex which were administered the vehicle only for 104 weeks. All surviving rats and mice were killed at 104 weeks.

Mean body weights of the male and female rats were lower in the dosed groups than in the corresponding control groups, and the depressions in weight were dose related. At the termination of the administration of the test chemical to the high-dose groups of rats, the mean body weights of these groups recovered rapidly. The mean body weights of the male mice were unaffected by the administration of the chemical; those of the females were only slightly affected. Mortality was generally higher in the dosed groups of rats and mice than in the corresponding control groups and was dose related in all tests except those using the female mice; however, sufficient numbers of animals of each species and sex were at risk for the development of late-appearing tumors.

In rats, proliferative squamous-cell lesions of the forestomach were observed in the dosed males (carcinomas: high-dose 1/50; papillomas: low-dose 1/47, high-dose 2/50; hyperplasias: low-dose 1/47, high-dose 2/50) and the dosed females (carcinomas: high-dose 1/48), but not in the male or female vehicle controls. The results of the Fisher exact test were not significant for squamous-cell papillomas or carcinomas. However, comparison of the incidence of these tumors in the high-dose males with that in 99 historical vehicle controls shows that the probability that three or more such tumors did not occur by chance, given that

none have been observed in the controls in this laboratory, is P = 0.014.

In mice, squamous-cell papillomas or carcinomas of the forestomach occurred in the low- and high-dose groups of each sex, but not in the corresponding control groups. The incidence in the high-dose males was significantly higher (P = 0.025) than that in the control males (males: vehicle controls 0/19, low-dose 2/43, high-dose 10/47 [21%]; females: vehicle controls 0/19, low-dose 1/45, high-dose 5/48 [10%]). Comparison of the incidences of these tumors in the high-dose males and females with those observed in the corresponding groups of 100 historical vehicle controls of each sex shows that the probability that their occurrence was not due to chance is P < 0.001. Also, a life- table analysis of the incidence in males indicated a significant (P = 0.003) increase in tumors over the period of observation (58 weeks to 104 weeks) in relation to an increase in dose.

Although the incidence of squamous-cell papillomas and carcinomas in male and female rats was significant only in males compared with historical vehicle controls, these tumors are of the same type as those appearing at the same site in male and female mice. Because these tumors are rare and not found in controls, and because they were found in dosed animals of both species, they are considered to be related to administration of the test chemical by gavage.

It is concluded that under the conditions of this bioassay, 3-(chloromethyl)pyridine hydrochloride was carcinogenic in male Fischer 344 rats and in B6C3F1 mice of both sexes, producing papillomas and carcinomas at the site of topical application, the stomach.

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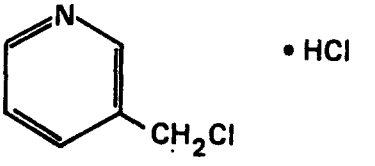
I. INTRODUCTION

3-(Chloromethyl)pyridine hydrochloride (CAS 6959-48-4; NCI C03838) is an intermediate that has been proposed for use in the synthesis of agricultural, pharmaceutical, and veterinary chemicals (Arnall and Clark, 1966). Its synthesis was achieved in the mid 1960's by the Midland Tar Distillers, Ltd., in Great Britain, through a process in which the alpha carbon of the alkyl side chain to the pyridine ring is chlorinated preferentially (Arnall and Clark, 1966). This discovery made it economically feasible for the first time to synthesize large quantities of monochloroalkylpyridine intermediates. This compound is neither manufactured in the United States, nor imported, at the present time (Stanford Research Institute, 1977; USITC, 1977a and 1977b), however, at the time it was selected for bioassay, it was felt that it could become a widely used industrial intermediate.



3(chloromethyl)pyridine (hydrochloride)

II. MATERIALS AND METHODS

A. Chemical

Three batches of the test chemical, hereinafter referred to as 3-(chloromethyl)pyridine hydrochloride, were obtained from Columbia Organic Chemicals, Columbia, South Carolina, for these studies. These batches were identified by the date of receipt at Midwest Research Institute, Kansas City, Missouri, as Lot No. C02-7-73, Lot No. C012-5-73, and Lot No. C02-25-75. All batches were used during the chronic studies; Lot No. C02-7-73 was also used during the subchronic studies.

The identity of each batch was confirmed by infrared, nuclear magnetic resonance, and ultraviolet spectral analyses. Elemental analyses (C, H, N, Cl) for $C_6H_7NCl_2$ were slightly low for chlorine in Lot Nos. C02-7-73 and C02-25-75. Trace impurities were found in all three lots by thin-layer chromatography. High-pressure liquid chromatography (ultraviolet detector, 254nm) indicated the presence of a single impurity, accounting for 0.2% of the total peak area in Lot No. C02-7-73, and of three minor impurities (0.49%) in Lot No. C02-25-75. Lot No. C02-7-73 contained 0.69 $\pm$ 0.04% water, Lot No. C012-5-73, $<$ 0.17%, and Lot No. C02-25-75, 0.82 $\pm$ 0.11%, as determined by Karl Fischer

analysis. Throughout this report the term used to represent this material is 3-(chloromethyl)pyridine hydrochloride.

These batches were stored at 4°C in the original containers.

B. Dosage Preparation

Solutions of 3-(chloromethyl)pyridine hydrochloride were prepared in distilled water (Borden Polar Water Co., Beltsville, Md.) at concentrations of 1 and 2% for mice and 0.75 and 1.50% for rats. These were administered by gavage on the same day on which they were prepared.

C. Animals

Fischer 344 rats and B6C3F1 mice of each sex were obtained from Charles River Breeding Laboratories, Wilmington, Massachusetts, under a contract with the Division of Cancer Treatment, NCI.

The animals were 28 days of age when received at the laboratory and were quarantined for 2 weeks prior to the start of the bioassay. Animals with clinical signs of disease and runts were killed. The remaining animals were segregated into equal weight groups and assigned to control or dosed groups in such a way that the mean weights of animals in each cage within a particular group were approximately the same.

D. Animal Maintenance

All animals were housed in temperature- and humidity-controlled rooms. The temperature was maintained at 21-25°C and the relative humidity at 45-55%. There were 10 changes of room air per hour, and the incoming and exhaust air was filtered through high efficiency particulate air (HEPA) filters (Flanders Filters, McLean, Va.). The animal rooms were positively pressurized with respect to the exit hall and negatively pressurized with respect to the entrance hall. Rooms were illuminated by cool white fluorescent lighting 8 hours per day.

Rats were housed four per cage and mice five per cage in solid polycarbonate cages (Lab Products, Inc., Garfield, N. J.). Each cage was covered with a wire mesh screen and a sheet of filter paper and contained heat-treated hardwood chip bedding (Absorb-Dri®, Lab Products, Garfield, N.J.) in the bottom. Cages and water bottles were sanitized two times per week, racks were washed each month, and feed hoppers were sanitized once per week at approximately 82°C; bedding was replaced two times per week and filter paper was replaced each month.

The animals were fed Wayne® Lab Blox (Allied Mills, Inc., Chicago, Ill.) and received fresh diets three times per week.

Water bottles contained tap water which had been acidified to pH 2.5.

Rats and mice were housed in separate rooms. Control and dosed animals were housed in the same room. Animals administered 3-(chloromethyl)pyridine hydrochloride were housed in the same room with animals administered the following chemicals:

Rats

Feed Studies

(CAS 105-55-5) N,N-diethylthiourea
(CAS 99-56-9) 4-nitro-o-phenylenediamine
(CAS 89-25-8) 1-phenyl-3-methyl-5-pyrazolone

Mice

Feed Studies

(CAS 2735-04-8) 2,4-dimethoxyaniline
(CAS 140-49-8) 4-chloroacetylacetanilide
(CAS 139-94-6) nithiazide
(CAS 624-18-0) p-phenylenediamine dihydrochloride
(CAS 99-56-9) 4-nitro-o-phenylenediamine
(CAS 89-25-8) 1-phenyl-3-methyl-5-pyrazolone

Gavage Studies

(CAS 512-56-1) trimethylphosphate
(CAS 4377-33-7) 2-(chloromethyl)pyridine hydrochloride
(CAS 1955-45-9) pivalolactone

E. Subchronic Studies

The LD_{50} for 3-(chloromethyl)pyridine hydrochloride administered orally to either Fischer 344 rats or B6C3F1 mice has been reported as 316 mg/kg (Litton-Bionetics, Inc., 1973).

Subchronic studies were conducted with Fischer 344 rats and B6C3F1 mice to estimate the maximum tolerated doses of 3-(chloromethyl)pyridine hydrochloride, on the basis of which two doses (hereinafter called "low" and "high" doses) were determined for the chronic studies. For the subchronic studies, the test chemical was administered by gavage, three times per week, at doses of 68, 100, 147, 215, or 316 mg/kg to rats, and 100, 147, 215, 316, or 464 mg/kg to mice. Five males and five females were tested at each dose, and groups of equal size served as vehicle controls, receiving distilled water only. Following a 7-week period of administration of the test chemical, the animals were observed for 1 week and then killed and necropsied.

Within the first 3 weeks of the subchronic test, two of the male rats and all of the female rats died at the highest dose, 316 mg/kg. In those male rats that were alive at week 7, mean body weights were depressed to the same extent in all groups, and in no case did this exceed 15% of controls. In the surviving females, there were no apparent effects on body weights.

In the mice, all males and females administered the highest dose, 464 mg/kg, died during the first week. At the end of week 7, there were only small weight depressions in the surviving female groups and no apparent weight depression in the males.

No signs of toxicity were found on gross pathologic examination of the organs taken from rats and mice.

The low and high doses for the chronic studies using rats were set at 75 and 150 mg/kg, and those for the chronic studies using mice were set at 100 and 200 mg/kg, respectively.

F. Chronic Studies

The test groups, doses administered, and times on study of the chronic studies are shown in tables 1 and 2.

G. Clinical and Pathologic Examinations

All animals were observed twice daily for signs of toxicity and were weighed and palpated for masses at regular intervals. Animals that were moribund and those that survived to the termination of the bioassay were killed using CO_2 and necropsied.

The pathologic evaluation consisted of gross and microscopic examination of major tissues, major organs, and all gross lesions from killed animals and from animals found dead. The following tissues were examined microscopically: skin, lymph nodes, mammary gland, salivary gland, bone marrow, trachea, lungs and bronchi, heart, thyroid, parathyroid, esophagus, stomach, small intestine, large intestine, liver, gallbladder (mice), pancreas, spleen, kidney, adrenal, urinary bladder, prostate or uterus, testis or

Table 1. Chronic Studies of 3-(Chloromethyl)pyridine Hydrochloride in Rats

Sex and Test Group	Initial No. of Animals[a]	3-(Chloromethyl) pyridine Hydrochloride Dose[b] (mg/kg)	Time on Study	
			Dosed (weeks)	Observed (weeks)
Male				
Vehicle-Control[c]	20	0		104
Low-Dose	50	75	103	1
High-Dose	50	150	83[d]	21
Female				
Vehicle-Control[c]	20	0		104
Low-Dose	50	75	103	1
High-Dose	50	150	83[d]	21

[a]Rats were approximately 6 weeks of age when placed on study.

[b]3-(Chloromethyl)pyridine hydrochloride was administered three times per week in distilled water at a volume of 1 ml per 100 gm body weight. Doses were calculated using the mean body weight of the group and were adjusted every month.

[c]Vehicle controls received distilled water three times per week at a volume of 1 ml per 100 gm body weight.

[d]Because of early deaths, the high-dose rats were dosed for only 83 weeks.

Table 2. Chronic Studies of 3-(Chloromethyl)pyridine Hydrochloride in Mice

Sex and Test Group	Initial No. of Animals[a]	3-(Chloromethyl) pyridine Hydrochloride Dose[b] (mg/kg)	Time on Study	
			Dosed (weeks)	Observed (weeks)
Male				
Vehicle-Control[c]	20	0		104
Low-Dose	50	100	102	2
High-Dose	50	200	81[d]	23
Female				
Vehicle-Control[c]	20	0		104
Low-Dose	50	100	102	2
High-Dose	50	200	81[d]	23

[a]Mice were approximately 6 weeks of age when placed on study.

[b]3-(Chloromethyl)pyridine hydrochloride was administered three times per week in distilled water at a volume of 1 ml per 100 gm body weight. Doses were calculated using the mean body weight of the group and were adjusted every month.

[c]Vehicle controls received distilled water three times per week at a volume of 1 ml per 100 gm body weight.

[d]Because of early deaths, the high-dose mice were dosed for only 81 weeks.

ovary, brain, and pituitary. Occasionally, additional tissues were also examined microscopically. The different tissues were preserved in 10% buffered formalin, embedded in paraffin, sectioned, and stained with hematoxylin and eosin. Special staining techniques were utilized when indicated for more definitive diagnosis.

A few tissues from some animals were not examined, particularly from those animals that died early. Also, some animals may have been missing, cannibalized, or judged to be in such an advanced state of autolysis as to preclude histopathologic evaluation. Thus, the number of animals from which particular organs or tissues were examined microscopically varies, and does not necessarily represent the number of animals that were placed on study in each group.

H. Data Recording and Statistical Analyses

Pertinent data on this experiment have been recorded in an automatic data processing system, the Carcinogenesis Bioassay Data System (Linhart et al., 1974). The data elements include descriptive information on the chemicals, animals, experimental design, clinical observations, survival, body weight, and individual pathologic results, as recommended by the International Union Against Cancer (Berenblum, 1969). Data tables were generated for

verification of data transcription and for statistical review.

These data were analyzed using the statistical techniques described in this section. Those analyses of the experimental results that bear on the possibility of carcinogenicity are discussed in the statistical narrative sections.

Probabilities of survival were estimated by the product-limit procedure of Kaplan and Meier (1958) and are presented in this report in the form of graphs. Animals were statistically censored as of the time that they died of other than natural causes or were found to be missing; animals dying from natural causes were not statistically censored. Statistical analyses for a possible dose-related effect on survival used the method of Cox (1972) for testing two groups for equality and Tarone's (1975) extensions of Cox's methods for testing for a dose-related trend. One-tailed P values have been reported for all tests except the departure from linearity test, which is only reported when its two-tailed P value is less than 0.05.

The incidence of neoplastic or nonneoplastic lesions has been given as the ratio of the number of animals bearing such lesions at a specific anatomic site (numerator) to the number of animals in which that site is examined (denominator). In most instances, the denominators included only those animals for which that site

was examined histologically. However, when macroscopic examination was required to detect lesions prior to histologic sampling (e.g., skin or mammary tumors), or when lesions could have appeared at multiple sites (e.g., lymphomas), the denominators consist of the numbers of animals necropsied.

The purpose of the statistical analyses of tumor incidence is to determine whether animals receiving the test chemical developed a significantly higher proportion of tumors than did the control animals. As a part of these analyses, the one-tailed Fisher exact test (Cox, 1970) was used to compare the tumor incidence of a control group with that of a group of dosed animals at each dose level. When results for a number of dosed groups (k) are compared simultaneously with those for a control group, a correction to ensure an overall significance level of 0.05 may be made. The Bonferroni inequality (Miller, 1966) requires that the P value for any comparison be less than or equal to 0.05/k. In cases where this correction was used, it is discussed in the narrative section. It is not, however, presented in the tables, where the Fisher exact P values are shown.

The Cochran-Armitage test for linear trend in proportions, with continuity correction (Armitage, 1971), was also used when appropriate. Under the assumption of a linear trend, this test determines if the slope of the dose-response curve is different

from zero at the one-tailed 0.05 level of significance. Unless otherwise noted, the direction of the significant trend is a positive dose relationship. This method also provides a two-tailed test of departure from linear trend.

A time-adjusted analysis was applied when numerous early deaths resulted from causes that were not associated with the formation of tumors. In this analysis, deaths that occurred before the first tumor was observed were excluded by basing the statistical tests on animals that survived at least 52 weeks, unless a tumor was found at the anatomic site of interest before week 52. When such an early tumor was found, comparisons were based exclusively on animals that survived at least as long as the animal in which the first tumor was found. Once this reduced set of data was obtained, the standard procedures for analyses of the incidence of tumors (Fisher exact tests, Cochran-Armitage tests, etc.) were followed.

When appropriate, life-table methods were used to analyze the incidence of tumors. Curves of the proportions surviving without an observed tumor were computed as in Saffiotti et al. (1972). The week during which an animal died naturally or was sacrificed was entered as the time point of tumor observation. Cox's methods of comparing these curves were used for two groups; Tarone's extension to testing for linear trend was used for three

groups. The statistical tests for the incidence of tumors which used life-table methods were one-tailed and, unless otherwise noted, in the direction of a positive dose relationship. Significant departures from linearity ($P < 0.05$, two-tailed test) were also noted.

The approximate 95 percent confidence interval for the relative risk of each dosed group compared to its control was calculated from the exact interval on the odds ratio (Gart, 1971). The relative risk is defined as p_t/p_c where p_t is the true binomial probability of the incidence of a specific type of tumor in a dosed group of animals and p_c is the true probability of the spontaneous incidence of the same type of tumor in a control group. The hypothesis of equality between the true proportion of a specific tumor in a dosed group and the proportion in a control group corresponds to a relative risk of unity. Values in excess of unity represent the condition of a larger proportion in the dosed group than in the control.

The lower and upper limits of the confidence interval of the relative risk have been included in the tables of statistical analyses. The interpretation of the limits is that in approximately 95% of a large number of identical experiments, the true ratio of the risk in a dosed group of animals to that in a control group would be within the interval calculated from the

experiment. When the lower limit of the confidence interval is greater than one, it can be inferred that a statistically significant result ($P < 0.025$ one-tailed test when the control incidence is not zero, $P < 0.050$ when the control incidence is zero) has occurred. When the lower limit is less than unity, but the upper limit is greater than unity, the lower limit indicates the absence of a significant result while the upper limit indicates that there is a theoretical possibility of the induction of tumors by the test chemical, which could not be detected under the conditions of this test.

III. RESULTS - RATS

A. Body Weights and Clinical Signs (Rats)

The mean body weights of the male and female rats were lower in the dosed groups than in the corresponding control groups, and the depressions in weight were dose related (figure 1). At the termination of the administration of the test chemical to the high-dose groups of rats (83 weeks), the mean body weights of these groups increased. Fluctuation in the growth curve may be due to mortality; as the size of a group diminishes, the mean body weight may be subject to variation. No other clinical signs related to administration of the test chemical were reported.

B. Survival (Rats)

The Kaplan and Meier curves estimating the probabilities of survival for male and female rats administered 3-(chloromethyl)pyridine hydrochloride by gavage at the doses of this bioassay, together with those of the vehicle controls, are shown in figure 2. The result of the Tarone test for positive dose-related trend in mortality is significant ($P < 0.001$) in each sex.

In male rats, 21/50 (42%) of the high-dose group, 38/50 (76%) of the low-dose group, and 19/20 (95%) of the control group survived

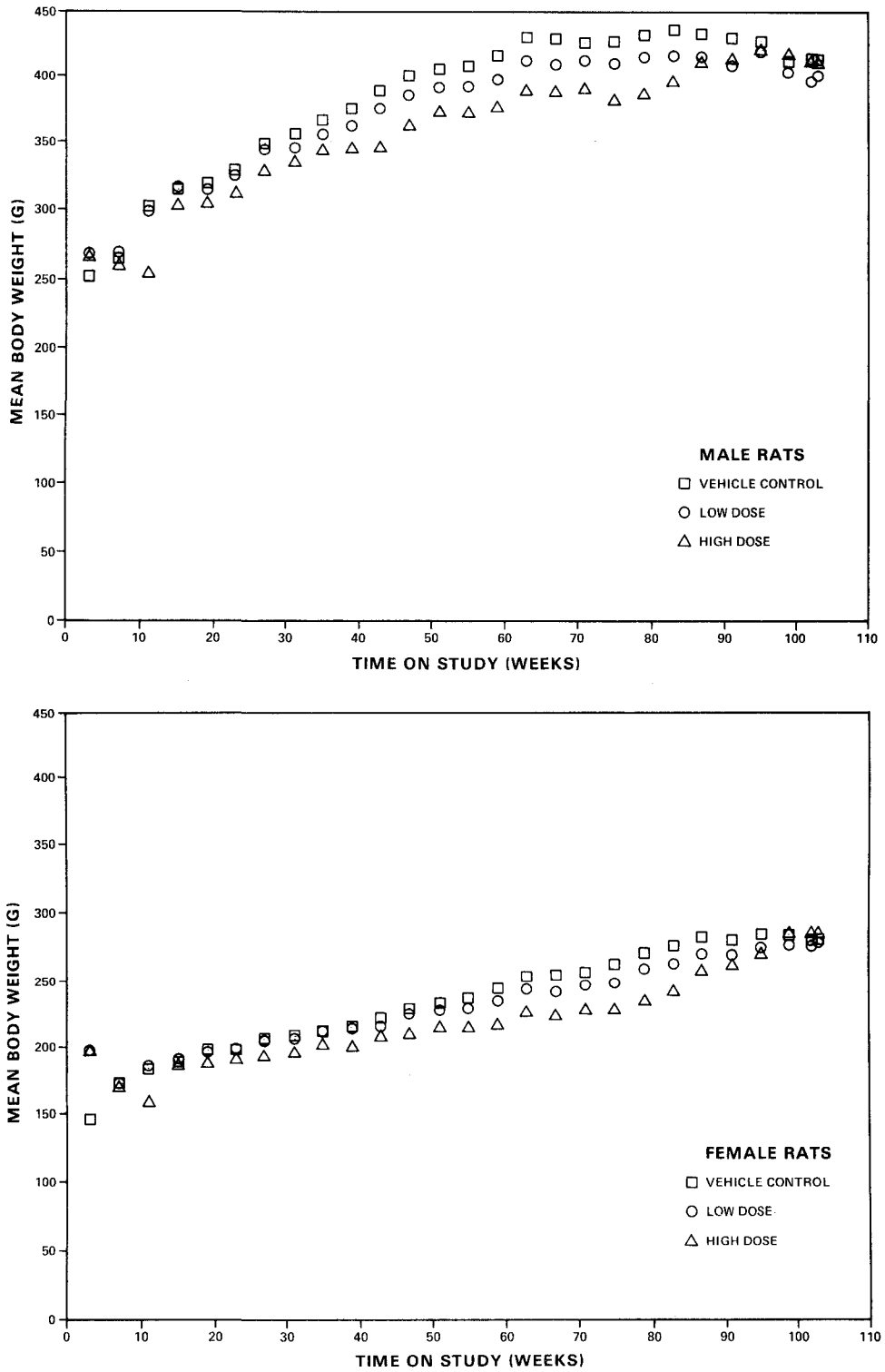


Figure 1. Growth Curves for Rats Administered 3-(Chloromethyl)Pyridine Hydrochloride by Gavage

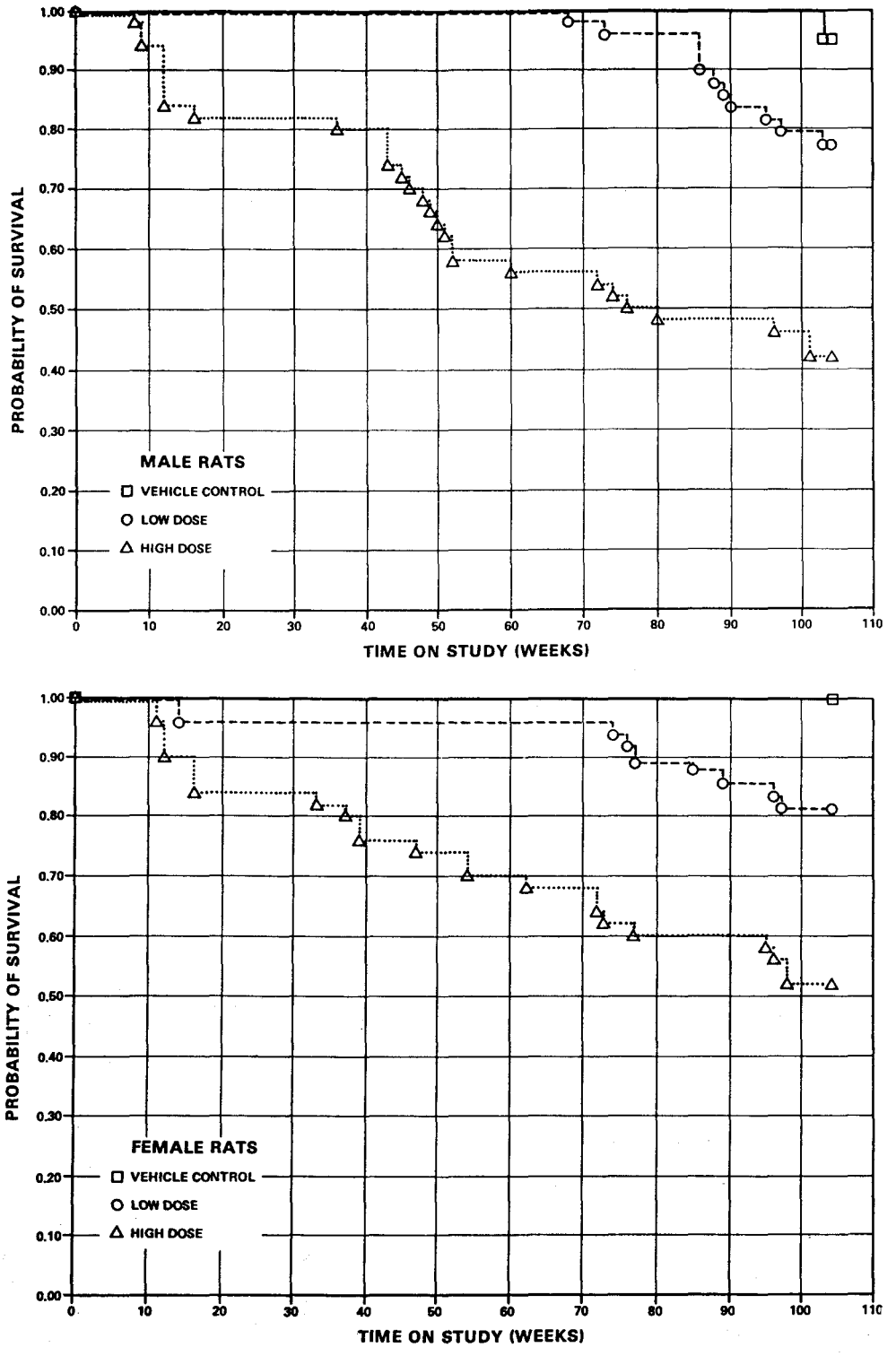


Figure 2. Survival Curves for Rats Administered 3-(Chloromethyl)Pyridine Hydrochloride by Gavage

to termination of the study. In females, 26/50 (52%) of the high-dose group, 40/50 (80%) of the low-dose group, and all 20 of the control group lived to termination of the study.

Sufficient numbers of rats of each sex were at risk for the development of late-appearing tumors.

C. Pathology (Rats)

Histopathologic findings on neoplasms in rats are summarized in Appendix A, tables A1 and A2; findings on nonneoplastic lesions are summarized in Appendix C, tables C1 and C2.

A variety of neoplastic lesions were evident in the control and dosed rats. Except for those of the forestomach, the tumors occurred in a random fashion in all groups. In the stomachs of dosed rats, there were both neoplastic and hyperplastic lesions as noted in the following table:

	Male Rats			Female Rats		
	Vehicle Control	Low Dose	High Dose	Vehicle Control	Low Dose	High Dose
Number of Animals with Stomach Examined Microscopically	19	47	50	20	45	48
Squamous-cell carcinoma			1(2%)			1(2%)
Squamous-cell papilloma		1(2%)	2(4%)			
Squamous-cell hyperplasia		1(2%)	2(4%)			

Grossly, the gastric nodules were described as single, white, pinpoint to 1 mm or 2 mm in diameter, warty growths, limited to the squamous portion of the stomach. Microscopically, the squamous-cell carcinomas were well differentiated and consisted of nests of squamous cells, some with keratin formation, that obliterated or invaded the muscularis mucosa but did not extend below the submucosa. The papillomas consisted of raised areas of spikes of hyperplastic squamous epithelium with hyperkeratotic caps. Chronic inflammation occurred in the submucosa beneath some of these lesions.

In organs other than the stomach, there were some degenerative and inflammatory lesions of the type usually encountered in aged rats, but none of the lesions were attributed to the test chemical. Chemically related lesions were not found in rats dying early in the study.

Based on the histopathologic examination, it was concluded that the gastric squamous-cell neoplastic lesions may be associated with the administration of 3-(chloromethyl)pyridine hydrochloride in rats under the conditions of this bioassay.

D. Statistical Analyses of Results (Rats)

Tables E1 and E2 in Appendix E contain the statistical analyses of the incidences of those primary tumors that occurred in at

least two animals of one group and at an incidence of at least 5% in one or more than one group. The early deaths in the high-dose group resulted in a shortened dosage period which obscures the meaning of trend analyses. Thus, these analyses have been omitted in the tables.

The results of the Fisher exact tests comparing the incidences of tumors in each of the dosed groups with that in the control group are not significant in the positive direction in either sex. The combined incidence of squamous-cell papillomas or carcinomas of the stomach in male rats was 0/19 in the controls, 1/47 (2%) in the low-dose group, and 3/50 (6%) in the high-dose group. The records of control animals at this laboratory indicate no such tumors occurred in 99 historical gavage vehicle-control male rats. The results of the Fisher exact test of these incidences are not significant, but under the estimate of 1% incidence in male control rats, the binomial probability (Fears, 1977) of three or more such tumors in 50 male rats is significant ($P = 0.014$). This analysis suggests an association between the occurrence of squamous-cell tumors in the high-dose group and the administration of the chemical. There was one squamous-cell carcinoma in the high-dose group of female rats compared with none in the 100 gavage vehicle-control female rats seen at this laboratory.

The results of the Fisher exact test on incidences of tumors of the pancreas and tumors of the testis in dosed male rats were significant in the negative direction. The higher incidences of tumors in the control group than in the dosed groups may have occurred because the dosed animals did not live as long as the control animals.

In each of the 95% confidence intervals of relative risk, shown in the tables, the value of one or less than one is included; this indicates the absence of significant positive results. It should also be noted that each of the intervals has an upper limit greater than one (except those of the incidences of islet-cell tumors and of tumors of the testis in high-dose male rats), indicating the theoretical possibility of the induction of tumors by 3-(chloromethyl)pyridine hydrochloride, which could not be detected under the conditions of this test.

IV. RESULTS - MICE

A. Body Weights and Clinical Signs (Mice)

The mean body weights of the male mice were unaffected by administration of the 3-(chloromethyl)pyridine hydrochloride, and those of the females were only slightly affected (figure 3). Fluctuation in the growth curve may be due to mortality; as the size of a group diminishes, the mean body weight may be subject to variation. No other clinical signs related to administration of the test chemical were reported.

B. Survival (Mice)

The Kaplan and Meier curves estimating the probabilities of survival for male and female mice administered 3-(chloromethyl)pyridine hydrochloride by gavage at the doses of this bioassay, together with those of the vehicle controls, are shown in figure 4. In male mice, the result of the Tarone test for positive dose-related trend in mortality is significant (P = 0.006). In females, the result of the Tarone test is not significant.

There were 23/50 (46%) of the male high-dose group, 30/50 (60%) of the low-dose group, and 15/20 (75%) of the control group surviving to termination of the study. There were 30/50 (60%) of

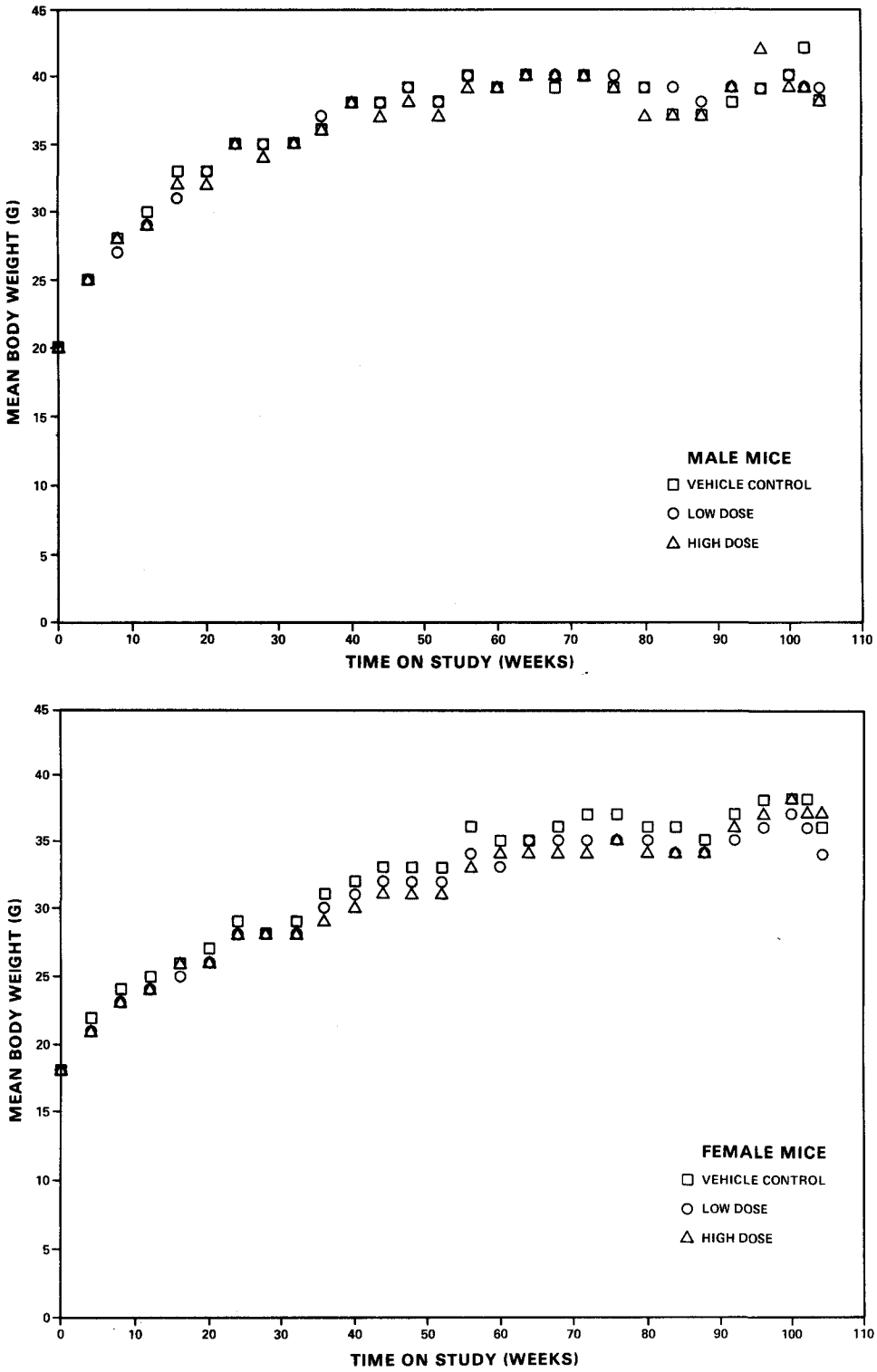
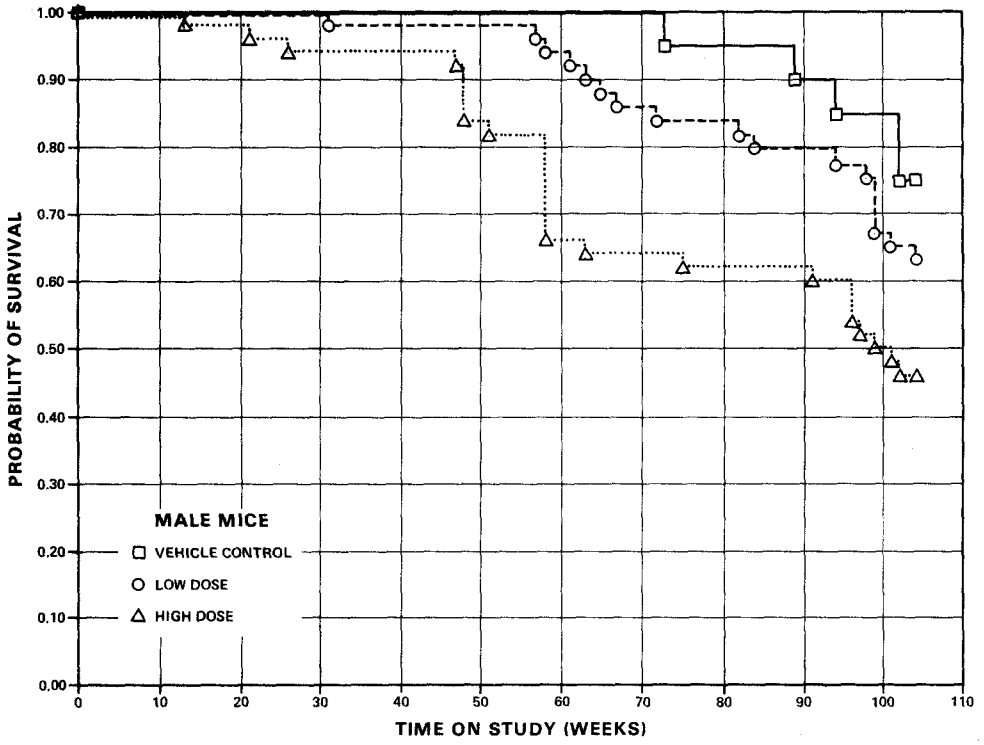


Figure 3. Growth Curves for Mice Administered 3-(Chloromethyl)Pyridine Hydrochloride by Gavage



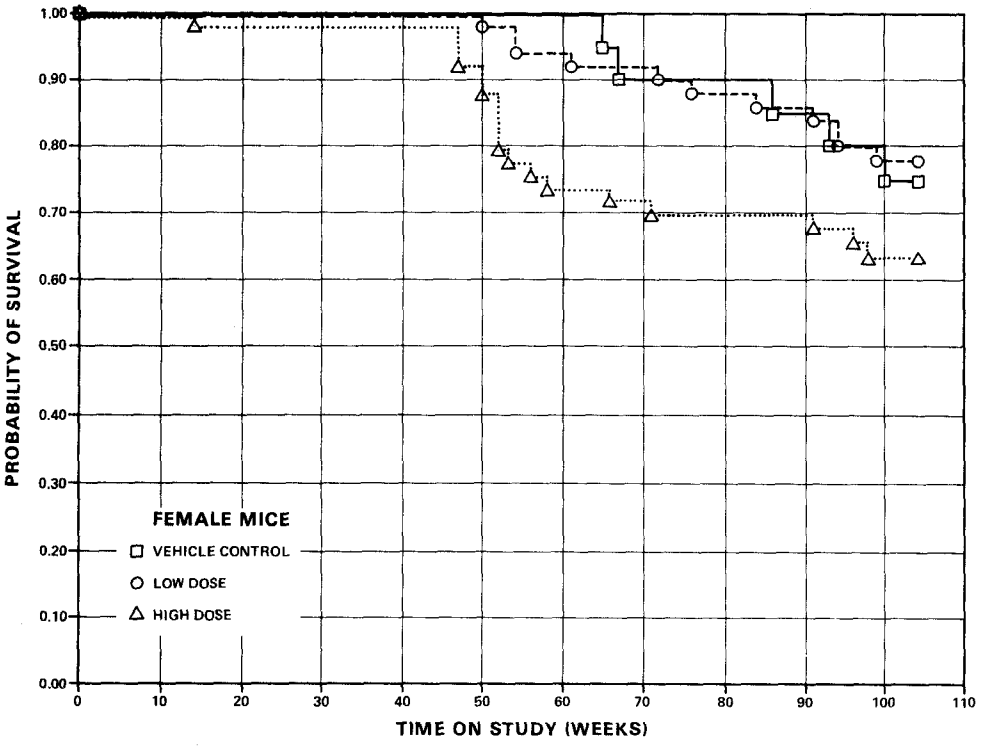


Figure 4. Survival Curves for Mice Administered 3-(Chloromethyl)Pyridine Hydrochloride by Gavage

the female high-dose group, 39/50 (78%) of the low-dose group, and 15/20 (75%) of the control group surviving to termination of the study.

Sufficient numbers of mice of each sex were at risk for the development of late-appearing tumors.

C. Pathology (Mice)

Histopathologic findings on neoplasms in mice are summarized in Appendix B, tables B1 and B2; findings on nonneoplastic lesions are summarized in Appendix D, tables D1 and D2.

A variety of neoplastic lesions were evident in control and dosed mice. Except for those in the stomach, they were spontaneous tumors occurring in a random fashion.

In the stomach of dosed mice, there were hyperplastic and neoplastic lesions that did not occur in the controls. The lesions were limited to the squamous portion of the stomach (forestomach) and occurred at the following incidence:

	Male Mice			Female Mice		
	Vehicle Control	Low Dose	High Dose	Vehicle Control	Low Dose	High Dose
Number of Animals with Stomach Examined Microscopically	19	43	47	19	45	48
Squamous-cell carcinoma			2(4%)			2(4%)
Squamous-cell papilloma		2(5%)	8(17%)		1(2%)	3(6%)
Squamous-cell hyperplasia		1(2%)	2(4%)			3(6%)

One of the squamous-cell carcinomas in a high-dose male mouse was described grossly as a large, hard mass in the stomach. Microscopically, this tumor, which arose from the forestomach, had invaded the liver and pancreas and metastasized to the mesenteric lymph node. The other squamous-cell carcinomas were well differentiated and consisted of epithelial nests that invaded through the muscularis mucosa or into the stalk but did not extend beyond the gastric submucosa.

Grossly, the papillomas were described as single, pinpoint to small white nodules arising from the squamous gastric mucosa. Histologically, in favorable sections, they consisted of tall hyperplastic spikes of squamous epithelium with hyperkeratotic caps that lined connective tissue stalks derived from the stomach wall.

The hyperplastic areas consisted of increased layers of basophi-

lic squamous cells and hyperkeratosis. In some of the affected mice there was mild to moderate chronic inflammation in the submucosa underlying the papillomas and hyperplastic areas.

Other tumors observed were those commonly recorded as spontaneous neoplasms of mice. These included a few hepatocellular and pulmonary tumors and some hematopoietic neoplasms, all with a slightly elevated incidence in the dosed mice. The lowered survival rates of the dosed mice made it difficult to interpret the incidences of these tumors, particularly when the differences in incidences between the dosed and control groups were marginal.

Other nonneoplastic lesions were of the type usually found in aged mice, and none was attributed to the test chemical. Chemical-related lesions did not occur in mice dying early in the study.

Based on the histopathologic examination, it was concluded that the squamous-cell tumors of the forestomach were associated with the administration of 3-(chloromethyl)pyridine hydrochloride in B6C3F1 mice under the conditions of this bioassay.

D. Statistical Analyses of Results (Mice)

Tables F1 and F2 in Appendix F contain the statistical analyses of the incidences of those primary tumors that occurred in at

least two animals of one group and at an incidence of at least 5% in one or more than one group. The early deaths in the high-dose group resulted in a shortened dosage period which obscures the meaning of trend analyses. Thus, these analyses have been omitted in the tables.

The probability level of the Fisher exact test comparing the combined incidence of squamous-cell papillomas or carcinomas of the stomach in the high-dose group of male mice with that in the control group of male mice is at the upper limit (P = 0.025) of that required for significance by the Bonferroni inequality criterion for multiple comparisons. In females, the results of the Fisher exact test are not significant; however, these tumors occurred only in the dosed groups. No such tumors have been observed at this laboratory in any of 100 historical gavage vehicle-control male or female mice. Under the estimate of the binomial parameter (Fears, 1977) of an incidence of 1% in control mice, the binomial probability of 10/47 (21%) or higher incidence in the high-dose male mice and of 5/48 (10%) incidence or higher in the high-dose female mice is significant at a level less than P = 0.001.

In addition to the analysis described above, a life-table analysis was performed, utilizing the time at which a tumor was observed (see section H, p. 13) and based on the time-weighted

dose calculated over the 104 weeks of the bioassay. This analysis of the incidence in males (controls 0/19, low-dose 2/43 [5%], high-dose 10/47 [21%]) ranging from the first tumor observed in the high-dose group (58 weeks) to the end of the bioassay (104 weeks) indicated a significant ($P = 0.003$) increase in the observation of tumors over this time period in relation to increase in dose. These analyses indicate an association of squamous-cell papillomas or carcinomas of the stomach with the administration of the test chemical.

V. DISCUSSION

3-(Chloromethyl)pyridine hydrochloride was toxic for Fischer 344 rats and B6C3F1 mice inasmuch as mean body weights were depressed in the dosed rats and mortality was generally higher in both dosed rats and dosed mice than in corresponding control groups. The nature of the toxic effect could not be established histopathologically. The depression in mean body weight was dose related in both sexes of rats, and the mortality was dose related in rats and male mice. In female mice only the survival of the high-dose group was affected. Because of early deaths in the high-dose groups of both the rats and the mice, administration of the test chemical was terminated about 20 weeks earlier for these groups than for the low-dose groups of both species. Sufficient numbers of animals of each species and sex were at risk, however, for the development of late-appearing tumors.

In rats, proliferative squamous-cell lesions of the stomach were observed in the dosed males (carcinomas: high-dose 1/50; papillomas: low-dose 1/47, high-dose 2/50; hyperplasias: low-dose 1/47, high-dose 2/50) and a dosed female (carcinomas: high- dose 1/48), but not in the male or female vehicle controls. The results of the Fisher exact test were not significant for squamous-cell papillomas or carcinomas. However, comparison of the incidence of these tumors in the high-dose males with that in

99 historical gavage vehicle controls shows that the probability that three or more such tumors did not occur by chance, given that none have been observed in the controls in this laboratory, is 0.014.

In mice, squamous-cell papillomas or carcinomas of the stomach occurred in the low- and high-dose groups of each sex, but not in the corresponding control groups of the 100 historical controls of each sex. The incidence in the high-dose males was significantly higher ($P = 0.025$) than that in the control males (males: vehicle controls 0/19, low-dose 2/43, high-dose 10/47 [21%]; females: vehicle controls 0/19, low-dose 1/45, high-dose 5/48 [10%]). Comparison of the incidences of these tumors in the high-dose males and females with those observed in the corresponding groups of 100 historical vehicle controls of each sex shows that the probability that their occurrence was not due to chance is $P < 0.001$. Also, a life-table analysis of the incidence in males indicated a significant ($P = 0.003$) increase in tumors over the period of observation (58 weeks to 104 weeks) in relation to an increase in dose.

Although the incidence of squamous-cell papillomas and carcinomas in male rats was significant only in comparison with historical vehicle controls, these tumors are of the same types as those that appeared at the same site in male and female mice. Because

these tumors are rare and not found in controls, and because they were found in dosed animals of both species, they are considered to be related to administration of the test chemical by gavage in both species.

It is concluded that under the conditions of this bioassay, 3-(chloromethyl)pyridine hydrochloride was carcinogenic in male Fischer 344 rats and B6C3F1 mice of both sexes, producing papillomas and carcinomas of the forestomach. Neoplastic lesions related to chemical administration were restricted to the site of topical application, the stomach.

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APPENDIX A

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN RATS ADMINISTERED 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

TABLE A1.

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	50	50
ANIMALS NECROPSIED	20	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	49	50
INTEGUMENTARY SYSTEM			
*SUBCUT TISSUE	(20)	(50)	(50)
FIBROMA		1 (2%)	
FIBROSARCOMA		1 (2%)	2 (4%)
NEUROFIBROMA		1 (2%)	
RESPIRATORY SYSTEM			
#LUNG	(20)	(49)	(49)
ALVEOLAR/BRONCHIOLAR CARCINOMA	2 (10%)	3 (6%)	
C-CELL CARCINOMA, METASTATIC	1 (5%)		
FIBROSARCOMA, METASTATIC		1 (2%)	
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(20)	(50)	(50)
LEUKEMIA,NOS	1 (5%)		2 (4%)
UNDIFFERENTIATED LEUKEMIA	1 (5%)	1 (2%)	
CIRCULATORY SYSTEM			
*BLOOD VESSEL	(20)	(50)	(50)
C-CELL CARCINOMA, METASTATIC		1 (2%)	
DIGESTIVE SYSTEM			
#LIVER	(19)	(48)	(48)
NEOPLASTIC NODULE	1 (5%)		
HEPATOCELLULAR CARCINOMA		1 (2%)	
#STOMACH	(19)	(47)	(50)
SQUAMOUS CELL PAPILLOMA		1 (2%)	2 (4%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1. MALE RATS: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
SQUAMOUS CELL CARCINOMA			1 (2%)
URINARY SYSTEM			
NONE			
ENDOCRINE SYSTEM			
#PITUITARY	(19)	(42)	(41)
CHROMOPHOBE ADENOMA	1 (5%)	4 (10%)	1 (2%)
CHROMOPHOBE CARCINOMA		3 (7%)	1 (2%)
#ADRENAL	(19)	(49)	(49)
CORTICAL ADENOMA	1 (5%)		1 (2%)
CORTICAL CARCINOMA	1 (5%)	1 (2%)	
PHEOCHROMOCYTOMA	2 (11%)	2 (4%)	1 (2%)
#THYROID	(17)	(39)	(41)
FOLLICULAR-CELL ADENOMA		1 (3%)	
FOLLICULAR-CELL CARCINOMA	1 (6%)		
C-CELL ADENOMA		1 (3%)	1 (2%)
C-CELL CARCINOMA	1 (6%)	1 (3%)	
#PANCREATIC ISLETS	(20)	(48)	(49)
ISLET-CELL ADENOMA	3 (15%)	2 (4%)	
ISLET-CELL CARCINOMA		1 (2%)	
REPRODUCTIVE SYSTEM			
*PREPUTIAL GLAND	(20)	(50)	(50)
ADENOMA, NOS	1 (5%)		
#TESTIS	(19)	(46)	(47)
INTERSTITIAL-CELL TUMOR	17 (89%)	38 (83%)	20 (43%)
NERVOUS SYSTEM			
#BRAIN	(20)	(46)	(47)
GLIOBLASTOMA MULTIFORME			1 (2%)
SPECIAL SENSE ORGANS			
NONE			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1. MALE RATS: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
MUSCULOSKELETAL SYSTEM			
*MANDIBLE	(20)	(50)	(50)
FIBROSARCOMA			1 (2%)
BODY CAVITIES			
*MESENTERY	(20)	(50)	(50)
LIPOMA		1 (2%)	
*TUNICA VAGINALIS	(20)	(50)	(50)
MESOTHELIOMA, NOS		1 (2%)	
ALL OTHER SYSTEMS			
NONE			
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	20	50	50
NATURAL DEATH@	1	10	27
MORIBUND SACRIFICE		1	2
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED		1	
TERMINAL SACRIFICE	19	38	21
ANIMAL MISSING			

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1. MALE RATS: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	20	45	24
TOTAL PRIMARY TUMORS	33	65	34
TOTAL ANIMALS WITH BENIGN TUMORS	19	41	21
TOTAL BENIGN TUMORS	25	52	26
TOTAL ANIMALS WITH MALIGNANT TUMORS	7	11	8
TOTAL MALIGNANT TUMORS	7	12	8
TOTAL ANIMALS WITH SECONDARY TUMORS#	1	2	
TOTAL SECONDARY TUMORS	1	2	
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT	1	1	
TOTAL UNCERTAIN TUMORS	1	1	
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE A2.

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE RATS ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	@50	50
ANIMALS NECROPSIED	20	49	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	49	50
INTEGUMENTARY SYSTEM			
*SKIN	(20)	(49)	(50)
TRICHOEPITHELIOMA		1 (2%)	
SEBACEOUS ADENOCARCINOMA		1 (2%)	
KERATOACANTHOMA		1 (2%)	
FIBROMA		1 (2%)	
*SUBCUT TISSUE	(20)	(49)	(50)
LIPOMA		1 (2%)	
MIXED MESENCHYMAL TUMOR, MALIGNA			1 (2%)
RESPIRATORY SYSTEM			
#LUNG	(20)	(48)	(50)
ALVEOLAR/BRONCHIOLAR CARCINOMA	1 (5%)		1 (2%)
CORTICAL CARCINOMA, METASTATIC	1 (5%)		
HEMATOPOIETIC SYSTEM			
#BRAIN	(19)	(49)	(50)
MALIGNANT RETICULOSIS			1 (2%)
*MULTIPLE ORGANS	(20)	(49)	(50)
UNDIFFERENTIATED LEUKEMIA	1 (5%)	2 (4%)	2 (4%)
LYMPHOCYTIC LEUKEMIA		1 (2%)	
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#STOMACH	(20)	(45)	(48)
SQUAMOUS CELL CARCINOMA			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

@ 50 ANIMALS WERE INITIALLY IN THE STUDY, BUT ONE ANIMAL WAS FOUND TO BE A MALE IN A FEMALE GROUP.

TABLE A2. FEMALE RATS: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
URINARY SYSTEM			
#URINARY BLADDER	(16)	(40)	(35)
PAPILLOMA, NOS		1 (3%)	
ENDOCRINE SYSTEM			
#PITUITARY	(19)	(43)	(40)
CHROMOPHOBE ADENOMA	8 (42%)	11 (26%)	13 (33%)
CHROMOPHOBE CARCINOMA	1 (5%)	1 (2%)	1 (3%)
#ADRENAL	(19)	(49)	(50)
CORTICAL CARCINOMA	1 (5%)		
PHEOCHROMOCYTOMA		1 (2%)	2 (4%)
#THYROID	(17)	(45)	(38)
FOLLICULAR-CELL ADENOMA		1 (2%)	
C-CELL ADENOMA		1 (2%)	
C-CELL CARCINOMA			1 (3%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(20)	(49)	(50)
SARCOMA, NOS		1 (2%)	
FIBROADENOMA	2 (10%)	4 (8%)	1 (2%)
*PREPUTIAL GLAND	(20)	(49)	(50)
ADENOMA, NOS	2 (10%)	1 (2%)	
*VAGINA	(20)	(49)	(50)
LEIOMYOMA			1 (2%)
#UTERUS	(19)	(47)	(49)
PAPILLARY ADENOMA		1 (2%)	
SARCOMA, NOS		1 (2%)	
ENDOMETRIAL STROMAL POLYP	2 (11%)	8 (17%)	2 (4%)
HEMANGIOPERICYTOMA, NOS		1 (2%)	
#OVARY	(18)	(45)	(48)
PAPILLARY ADENOMA			1 (2%)
NERVOUS SYSTEM			
#BRAIN	(19)	(49)	(50)
CHROMOPHOBE CARCINOMA, INVASIVE	1 (5%)		

\# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2. FEMALE RATS: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
EPENDYMOMA		1 (2%)	
SPECIAL SENSE ORGANS			
NONE			
MUSCULOSKEIETAL SYSTEM			
NONE			
BODY CAVITIES			
*MEDIASTINUM	(20)	(49)	(50)
MESCTHELIOMA, NOS	1 (5%)		
*MESENTERY	(20)	(49)	(50)
LIPOMA			1 (2%)
ALL OTHER SYSTEMS			
SITE UNKNOWN			
LIPOMA		1	
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	20	50	50
NATURAL DEATH@		7	21
MORIBUND SACRIFICE		2	3
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	20	40	26
ANIMAL MISSING			
ANIMAL DELETED (WRONG SEX)		1	

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2. FEMALE RATS: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	14	31	23
TOTAL PRIMARY TUMORS	19	43	29
TOTAL ANIMALS WITH BENIGN TUMORS	12	26	18
TOTAL BENIGN TUMORS	14	34	21
TOTAL ANIMALS WITH MALIGNANT TUMORS	4	8	5
TOTAL MALIGNANT TUMORS	4	8	8
TOTAL ANIMALS WITH SECONDARY TUMORS#	2		
TOTAL SECONDARY TUMORS	2		
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT	1	1	
TOTAL UNCERTAIN TUMORS	1	1	
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

APPENDIX B

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MICE ADMINISTERED 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

TABLE B1.

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	50	50
ANIMALS MISSING		2	
ANIMALS NECROPSIED	20	48	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	48	50
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG	(20)	(45)	(48)
HEPATOCELLULAR CARCINOMA, METAST	1 (5%)		
ALVEOLAR/BRONCHIOLAR ADENOMA	1 (5%)		
ALVEOLAR/BRONCHIOLAR CARCINOMA	1 (5%)	4 (9%)	5 (10%)
OSTEOSARCOMA, METASTATIC	1 (5%)		
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(20)	(48)	(50)
MALIGNANT LYMPHOMA, NOS	1 (5%)	2 (4%)	2 (4%)
MALIGNANT LYMPHOMA, MIXED TYPE	1 (5%)	1 (2%)	1 (2%)
LEUKEMIA,NOS			1 (2%)
UNDIFFERENTIATED LEUKEMIA	1 (5%)	3 (6%)	3 (6%)
LYMPHOCYTIC LEUKEMIA		1 (2%)	1 (2%)
#BRONCHIAL LYMPH NODE	(16)	(33)	(35)
ALVEOLAR/BRONCHIOLAR CA, METASTA			1 (3%)
#MESENTERIC L. NODE	(16)	(33)	(35)
SQUAMOUS CELL CARCINOMA, METASTA			1 (3%)
MALIGNANT LYMPHOMA, NOS			1 (3%)
MALIGNANT LYMPHOMA, MIXED TYPE		2 (6%)	1 (3%)
CIRCULATORY SYSTEM			
*BLOOD VESSEL	(20)	(48)	(50)
ALVEOLAR/BRONCHIOLAR CA, METASTA		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B1. MALE MICE: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
DIGESTIVE SYSTEM			
#LIVER	(20)	(46)	(49)
SQUAMOUS CELL CARCINOMA, INVASIV			1 (2%)
HEPATOCELLULAR ADENOMA	1 (5%)		
HEPATOCELLULAR CARCINOMA	2 (10%)	5 (11%)	9 (18%)
#PANCREAS	(18)	(33)	(36)
SQUAMOUS CELL CARCINOMA, INVASIV			1 (3%)
#STOMACH	(19)	(43)	(47)
SQUAMOUS CELL PAPILLOMA		2 (5%)	8 (17%)
SQUAMOUS CELL CARCINOMA			2 (4%)
URINARY SYSTEM			
NONE			
ENDOCRINE SYSTEM			
#THYROID	(10)	(31)	(35)
FOLLICULAR-CELL CARCINOMA			1 (3%)
REPRODUCTIVE SYSTEM			
#TESTIS	(19)	(45)	(44)
INTERSTITIAL-CELL TUMOR		1 (2%)	
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
NONE			
MUSCULOSKELETAL SYSTEM			
NONE			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B1. MALE MICE: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
BODY CAVITIES			
NONE			
ALL OTHER SYSTEMS			
SITE UNKNOWN			
SARCOMA, NOS		1	
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	20	50	50
NATURAL DEATH@	4	17	25
MORIBUND SACRIFICE	1	1	2
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	15	30	23
ANIMAL MISSING		2	
@ INCLUDES AUTOLYZED ANIMALS			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	7	20	28
TOTAL PRIMARY TUMORS	8	22	35
TOTAL ANIMALS WITH BENIGN TUMORS	2	3	8
TOTAL BENIGN TUMORS	2	3	8
TOTAL ANIMALS WITH MALIGNANT TUMORS	5	19	24
TOTAL MALIGNANT TUMORS	6	19	27
TOTAL ANIMALS WITH SECONDARY TUMORS#	2	1	2
TOTAL SECONDARY TUMORS	2	1	4
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT			
TOTAL UNCERTAIN TUMORS			
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE B2.

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE MICE ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	50	50
ANIMALS MISSING			1
ANIMALS NECROPSIED	20	50	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	50	49
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG	(18)	(50)	(49)
HEPATOCELLULAR CARCINOMA, METAST			1 (2%)
ALVECLAR/BRONCHIOLAR ADENOMA		1 (2%)	
ALVEOLAR/BRONCHIOLAR CARCINOMA	1 (6%)		3 (6%)
OSTEOSARCOMA, METASTATIC		1 (2%)	
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(20)	(50)	(49)
MALIGNANT LYMPHOMA, NOS	1 (5%)	2 (4%)	1 (2%)
MALIG. LYMPHOMA, HISTIOCYTIC TYPE			1 (2%)
MALIGNANT LYMPHOMA, MIXED TYPE			2 (4%)
LEUKEMIA, NOS			1 (2%)
UNDIFFERENTIATED LEUKEMIA		2 (4%)	2 (4%)
LYMPHOCYTIC LEUKEMIA	1 (5%)	1 (2%)	
#LYMPH NODE	(16)	(42)	(40)
ALVECLAR/BRONCHIOLAR CA, METASTA			1 (3%)
#MANDIBULAR L. NODE	(16)	(42)	(40)
MALIGNANT LYMPHOMA, MIXED TYPE			1 (3%)
#MESENTERIC L. NODE	(16)	(42)	(40)
MALIGNANT LYMPHOMA, MIXED TYPE		4 (10%)	2 (5%)
CIRCULATORY SYSTEM			
*BLOOD VESSEL	(20)	(50)	(49)
FOLLICULAR-CELL CARCINOMA, INVAS		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2. FEMALE MICE: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
DIGESTIVE SYSTEM			
#LIVER	(20)	(50)	(49)
HEPATOCELLULAR CARCINOMA	1 (5%)		2 (4%)
#STOMACH	(19)	(45)	(48)
SQUAMOUS CELL PAPILLOMA		1 (2%)	3 (6%)
SQUAMOUS CELL CARCINOMA			2 (4%)
URINARY SYSTEM			
NONE			
ENDOCRINE SYSTEM			
#PITUITARY	(9)	(25)	(29)
CHROMOPHOBE ADENOMA			2 (7%)
#ADRENAL	(16)	(41)	(43)
CORTICAL ADENOMA		1 (2%)	
#THYROID	(8)	(15)	(36)
FOLLICULAR-CELL CARCINOMA		1 (7%)	
REPRODUCTIVE SYSTEM			
#UTERUS	(18)	(47)	(47)
ENDOMETRIAL STROMAL POLYP			1 (2%)
#OVARY	(6)	(13)	(33)
PAPILLARY ADENOMA		1 (8%)	
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
*EYE/LACRIMAL GLAND	(20)	(50)	(49)
ADENOCARCINOMA, NOS			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2. FEMALE MICE: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*ABDOMINAL CAVITY	(20)	(50)	(49)
SARCOMA, NOS	1 (5%)		
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(20)	(50)	(49)
SARCOMA, NOS		1 (2%)	
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	20	50	50
NATURAL DEATH@	4	11	17
MORIBUND SACRIFICE	1		2
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	15	39	30
ANIMAL MISSING			1

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2. FEMALE MICE: NEOPLASMS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	5	14	20
TOTAL PRIMARY TUMORS	5	15	24
TOTAL ANIMALS WITH BENIGN TUMORS		4	6
TOTAL BENIGN TUMORS		4	6
TOTAL ANIMALS WITH MALIGNANT TUMORS	5	11	17
TOTAL MALIGNANT TUMORS	5	11	18
TOTAL ANIMALS WITH SECONDARY TUMORS#		2	2
TOTAL SECONDARY TUMORS		2	2
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT			
TOTAL UNCERTAIN TUMORS			
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

APPENDIX C

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN RATS ADMINISTERED 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

TABLE C1.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	50	50
ANIMALS NECROPSIED	20	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	49	50
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG/BRONCHUS	(20)	(49)	(49)
POLYPOID HYPERPLASIA		1 (2%)	
#LUNG	(20)	(49)	(49)
CONGESTION, NOS			7 (14%)
EDEMA, NOS			1 (2%)
HEMORRHAGE			1 (2%)
PNEUMONIA, CHRONIC MURINE			1 (2%)
GRANULOMA, FOREIGN BODY		1 (2%)	
HYPERPLASIA, ADENOMATOUS		2 (4%)	1 (2%)
HEMATOPOIETIC SYSTEM			
#SPLEEN	(20)	(48)	(48)
INFARCT, NOS		1 (2%)	
HEMOSIDEROSIS			1 (2%)
HEMATOPOIESIS	1 (5%)		
#MESENTERIC L. NODE	(19)	(47)	(43)
HISTIOCYTOSIS		1 (2%)	
CIRCULATORY SYSTEM			
#HEART	(19)	(48)	(46)
CALCIFICATION, NOS			1 (2%)
#MYOCARDIUM	(19)	(48)	(46)
INFLAMMATION, FOCAL			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
INFLAMMATION, CHRONIC FOCAL		6 (13%)	2 (4%)
FIBROSIS	1 (5%)	3 (6%)	1 (2%)
FIBROSIS, FOCAL	2 (11%)	4 (8%)	3 (7%)
*BLOOD VESSEL	(20)	(50)	(50)
CALCIFICATION, NOS			1 (2%)
*ARTERY	(20)	(50)	(50)
INFLAMMATION, CHRONIC			1 (2%)
*AORTA	(20)	(50)	(50)
INFLAMMATION, CHRONIC		1 (2%)	
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(19)	(48)	(44)
INFLAMMATION, NOS			1 (2%)
FIBROSIS, DIFFUSE			1 (2%)
#LIVER	(19)	(48)	(48)
CONGESTION, PASSIVE			2 (4%)
INFLAMMATION, ACUTE FOCAL	1 (5%)		
INFLAMMATION, GRANULOMATOUS		1 (2%)	
CIRRHOSIS, NOS		1 (2%)	
METAMORPHOSIS FATTY	2 (11%)		
EOSINOPHILIC CYTO CHANGE	1 (5%)		
#HEPATIC CAPSULE	(19)	(48)	(48)
HYPERPLASIA, FOCAL		1 (2%)	
*BILE DUCT	(20)	(50)	(50)
HYPERPLASIA, NOS	17 (85%)	34 (68%)	22 (44%)
#PANCREAS	(20)	(48)	(49)
ATROPHY, NOS			1 (2%)
#PANCREATIC ACINUS	(20)	(48)	(49)
ATROPHY, NOS		1 (2%)	
ATROPHY, FOCAL		2 (4%)	
#ESOPHAGUS	(20)	(47)	(44)
INFLAMMATION, ACUTE		1 (2%)	
#STOMACH	(19)	(47)	(50)
DILATATION, NOS			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
HYPERPLASIA, EPITHELIAL		1 (2%)	2 (4%)
#GASTRIC MUCOSA	(19)	(47)	(50)
DILATATION, NOS	12 (63%)	23 (49%)	15 (30%)
#COLON	(20)	(47)	(48)
PARASITISM	14 (70%)	18 (38%)	10 (21%)
URINARY SYSTEM			
#KIDNEY	(20)	(49)	(49)
CONGESTION, NOS			6 (12%)
GLOMERULONEPHRITIS, NOS	12 (60%)	28 (57%)	13 (27%)
INFLAMMATION, CHRONIC			1 (2%)
NEPHROSIS, HEMOGLOBINURIC			1 (2%)
CALCIFICATION, NOS			1 (2%)
#KIDNEY/TUBULE	(20)	(49)	(49)
NECROSIS, NOS			1 (2%)
NECROSIS, FOCAL	1 (5%)		
ENDOCRINE SYSTEM			
#PITUITARY	(19)	(42)	(41)
CYST, NOS		1 (2%)	
HYPERPLASIA, CHROMOPHOBE-CELL		2 (5%)	1 (2%)
#ADRENAL	(19)	(49)	(49)
CYTOPLASMIC VACUOLIZATION	1 (5%)	1 (2%)	2 (4%)
HYPERPLASIA, NODULAR			1 (2%)
ANGIECTASIS		6 (12%)	2 (4%)
#THYROID	(17)	(39)	(41)
CYSTIC FOLLICLES		1 (3%)	1 (2%)
INFLAMMATION, CHRONIC FOCAL			1 (2%)
HYPERPLASIA, C-CELL	2 (12%)		1 (2%)
#PARATHYROID	(8)	(19)	(17)
HYPERPLASIA, ADENOMATOUS	1 (13%)		
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(20)	(50)	(50)
CYSTIC DUCTS		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
#PROSTATE	(17)	(43)	(41)
INFLAMMATION, SUPPURATIVE			1 (2%)
INFLAMMATION, ACUTE FOCAL	1 (6%)		
HYPERPLASIA, NOS			1 (2%)
#TESTIS	(19)	(46)	(47)
TORSION		1 (2%)	
ATROPHY, NOS	2 (11%)	4 (9%)	1 (2%)
HYPERPLASIA, INTERSTITIAL CELL		1 (2%)	
#TESTIS/TUBULE	(19)	(46)	(47)
CALCIFICATION, NOS		1 (2%)	
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
NONE			
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*MESENTERY	(20)	(50)	(50)
PERIARTERITIS	1 (5%)		
NECROSIS, FAT			1 (2%)
ALL OTHER SYSTEMS			
NONE			
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED		1	10

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
AUTO/NECROPSY/HISTO PERF		2	1
AUTC/NECROPSY/NO HISTO		1	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	@50	50
ANIMALS NECROPSIED	20	49	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	49	50
INTEGUMENTARY SYSTEM			
*SUBCUT TISSUE	(20)	(49)	(50)
EPIDERMAL INCLUSION CYST		1 (2%)	
RESPIRATORY SYSTEM			
#TRACHEA	(17)	(44)	(41)
INFLAMMATION, CHRONIC		1 (2%)	
HYPERPLASIA, EPITHELIAL			1 (2%)
METAPLASIA, SQUAMOUS		1 (2%)	
#LUNG	(20)	(48)	(50)
CONGESTION, NOS			8 (16%)
ABSCESS, NOS		1 (2%)	
PNEUMONIA, CHRONIC MURINE	1 (5%)	1 (2%)	2 (4%)
INFLAMMATION, GRANULOMATOUS		1 (2%)	
GRANULOMA, NOS	1 (5%)		
GRANULOMA, FOREIGN BODY	1 (5%)	1 (2%)	
HYPERPLASIA, ADENOMATOUS		2 (4%)	
HYPERPLASIA, ALVEOLAR EPITHELIUM		1 (2%)	1 (2%)
HISTIOCYTOSIS		2 (4%)	1 (2%)
LEUKEMOID REACTION			1 (2%)
HEMATOPOIETIC SYSTEM			
*SPLEEN	(20)	(48)	(49)
HEMATOMA, ORGANIZED	1 (5%)		
FIBROSIS, FOCAL			1 (2%)
INFARCT, NOS		1 (2%)	
HEMOSIDEROSIS			1 (2%)
HEMATOPOIESIS		2 (4%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

@ 50 ANIMALS WERE INITIALLY IN THE STUDY, BUT ONE ANIMAL WAS FOUND TO BE A MALE IN A FEMALE GROUP.

TABLE C2. FEMALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
CIRCULATORY SYSTEM			
#MYOCARDIUM	(20)	(47)	(49)
INFLAMMATION, ACUTE FOCAL			1 (2%)
INFLAMMATION, CHRONIC FOCAL	4 (20%)	6 (13%)	8 (16%)
FIBROSIS, FOCAL	1 (5%)	1 (2%)	1 (2%)
DIGESTIVE SYSTEM			
#LIVER	(20)	(48)	(49)
CONGESTION, NOS			1 (2%)
CONGESTION, CHRONIC PASSIVE		1 (2%)	
INFLAMMATION, NECROTIZING		1 (2%)	
GRANULOMA, NOS		1 (2%)	
NECROSIS, FOCAL		1 (2%)	2 (4%)
METAMORPHOSIS FATTY		1 (2%)	
BASOPHILIC CYTO CHANGE			1 (2%)
*BILE DUCT	(20)	(49)	(50)
INFLAMMATION, CHRONIC		1 (2%)	
HYPERPLASIA, NOS	5 (25%)	21 (43%)	11 (22%)
#PANCREAS	(20)	(46)	(49)
INFLAMMATION, INTERSTITIAL		1 (2%)	
#PANCREATIC ACINUS	(20)	(46)	(49)
ATROPHY, NOS		1 (2%)	1 (2%)
ATROPHY, FOCAL	1 (5%)	1 (2%)	2 (4%)
#GASTRIC MUCOSA	(20)	(45)	(48)
DILATATION, NOS	12 (60%)	27 (60%)	23 (48%)
#COLON	(20)	(46)	(49)
PARASITISM	7 (35%)	17 (37%)	10 (20%)
URINARY SYSTEM			
#KIDNEY	(19)	(49)	(50)
CONGESTION, NOS			7 (14%)
GLOMERULONEPHRITIS, NOS	4 (21%)	4 (8%)	3 (6%)
#KIDNEY/CORTEX	(19)	(49)	(50)
CYST, NOS		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2. FEMALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
#KIDNEY/TUBULE	(19)	(49)	(50)
NECROSIS, NOS	1 (5%)		
NECROSIS, FOCAL		1 (2%)	
ENDOCRINE SYSTEM			
#PITUITARY	(19)	(43)	(40)
CYST, NOS	1 (5%)	4 (9%)	1 (3%)
NECROSIS, FOCAL		1 (2%)	
HYPERPLASIA, CHROMOPHOBE-CELL		1 (2%)	
ANGIECTASIS		1 (2%)	
#ADRENAL	(19)	(49)	(50)
CYTOPLASMIC VACUOLIZATION	1 (5%)	4 (8%)	3 (6%)
ANGIECTASIS		3 (6%)	
HEMATOPOIESIS			1 (2%)
#THYROID	(17)	(45)	(38)
INFLAMMATION, CHRONIC FOCAL	1 (6%)		
HYPERPLASIA, C-CELL		2 (4%)	3 (8%)
HYPERPLASIA, FOLLICULAR-CELL			1 (3%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(20)	(49)	(50)
CYST, NOS	1 (5%)		
CYSTIC DUCTS	1 (5%)		
INFLAMMATION, CHRONIC	1 (5%)		
HYPERPLASIA, NOS			1 (2%)
*MAMMARY DUCT	(20)	(49)	(50)
HYPERPLASIA, NOS			1 (2%)
#UTERUS	(19)	(47)	(49)
HYDROMETRA		1 (2%)	2 (4%)
#CERVIX UTERI	(19)	(47)	(49)
CYST, NOS	1 (5%)		
#UTERUS/ENDOMETRIUM	(19)	(47)	(49)
ABSCESS, NOS			1 (2%)
HYPERPLASIA, CYSTIC	1 (5%)	1 (2%)	1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2. FEMALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
#OVARY	(18)	(45)	(48)
CYST, NOS	1 (6%)	3 (7%)	2 (4%)
NERVOUS SYSTEM			
#BRAIN	(19)	(49)	(50)
HEMORRHAGE		1 (2%)	
SPECIAL SENSE ORGANS			
*EYE	(20)	(49)	(50)
HEMORRHAGE		1 (2%)	
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*ABDOMINAL CAVITY	(20)	(49)	(50)
ABSCESS, NOS			1 (2%)
NECROSIS, NOS			1 (2%)
*EPICARDIUM	(20)	(49)	(50)
INFLAMMATION, FIBRINOUS		1 (2%)	
ALL OTHER SYSTEMS			
SITE UNKNOWN			
NECROSIS, FAT		1	
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	1	2	8

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

APPENDIX D

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MICE ADMINISTERED 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

TABLE D1.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	50	50
ANIMALS MISSING		2	
ANIMALS NECROPSIED	20	48	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	48	50
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG	(20)	(45)	(48)
CONGESTION, NOS		2 (4%)	7 (15%)
BRONCHOPNEUMONIA, ACUTE	1 (5%)		
PNEUMONIA, CHRONIC MURINE	3 (15%)	2 (4%)	1 (2%)
PERIVASCULAR CUFFING	1 (5%)	1 (2%)	
HYPERPLASIA, ADENOMATOUS	1 (5%)		2 (4%)
HISTIOCYTOSIS			3 (6%)
LEUKEMOID REACTION			1 (2%)
HYPERPLASIA, BASOPHILIC		1 (2%)	
#LUNG/ALVEOLI	(20)	(45)	(48)
CRYSTALS, NOS			1 (2%)
HISTIOCYTOSIS	1 (5%)		
HEMATOPOIETIC SYSTEM			
#SPLEEN	(17)	(41)	(41)
AMYLOIDOSIS			1 (2%)
ANGIECTASIS		1 (2%)	
HEMATOPOIESIS		3 (7%)	1 (2%)
#MESENTERIC L. NODE	(16)	(33)	(35)
CONGESTION, NOS		1 (3%)	
HEMORRHAGE		4 (12%)	1 (3%)
HEMOSIDEROSIS		1 (3%)	1 (3%)
HISTIOCYTOSIS		1 (3%)	
MASTOCYTOSIS		1 (3%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1. MALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
CIRCULATORY SYSTEM			
*PORTAL VEIN	(20)	(48)	(50)
THROMBOSIS, NOS		1 (2%)	
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(19)	(39)	(44)
PERIVASCULAR CUFFING	1 (5%)	2 (5%)	2 (5%)
#LIVER	(20)	(46)	(49)
CYST, NOS	1 (5%)		
CONGESTION, NOS		1 (2%)	
HEMATOMA, NOS		1 (2%)	
INFLAMMATION, FOCAL	1 (5%)		
INFLAMMATION, CHRONIC	1 (5%)	1 (2%)	
INFLAMMATION, CHRONIC FOCAL	1 (5%)	4 (9%)	
GRANULATION, TISSUE		1 (2%)	
THROMBOPHLEBITIS			1 (2%)
FIBROSIS		1 (2%)	
PERIVASCULAR CUFFING	1 (5%)		
NECROSIS, FOCAL		1 (2%)	1 (2%)
INFARCT, NOS		2 (4%)	
CALCIUM DEPOSIT		1 (2%)	
CALCIFICATION, NOS		1 (2%)	
BASOPHILIC CYTO CHANGE	1 (5%)		
ATROPHY, NOS	1 (5%)		
#LIVER/HEPATOCYTES	(20)	(46)	(49)
CYTOPLASMIC CHANGE, NOS		1 (2%)	
#PANCREAS	(18)	(33)	(36)
INFLAMMATION, INTERSTITIAL			1 (3%)
#PANCREATIC DUCT	(18)	(33)	(36)
NECROSIS, NOS	1 (6%)		
#PANCREATIC ACINUS	(18)	(33)	(36)
NECROSIS, NOS	1 (6%)		
ATROPHY, NOS	1 (6%)		
#STOMACH	(19)	(43)	(47)
CYST, NOS			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1. MALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
INFLAMMATION, NOS			1 (2%)
HYPERPLASIA, EPITHELIAL		1 (2%)	2 (4%)
#PEYERS PATCH	(17)	(42)	(42)
HYPERPLASIA, NOS	1 (6%)	2 (5%)	
#COLON	(18)	(44)	(47)
PARASITISM	3 (17%)	12 (27%)	13 (28%)
URINARY SYSTEM			
#KIDNEY	(20)	(46)	(47)
CONGESTION, NOS		2 (4%)	7 (15%)
PERIVASCULAR CUFFING	3 (15%)	7 (15%)	5 (11%)
#KIDNEY/MEDULLA	(20)	(46)	(47)
CYST, NOS			1 (2%)
#PERIRENAL TISSUE	(20)	(46)	(47)
NECROSIS, FOCAL		1 (2%)	
#KIDNEY/TUBULE	(20)	(46)	(47)
HYPERPLASIA, FOCAL	1 (5%)		
#KIDNEY/PELVIS	(20)	(46)	(47)
INFLAMMATION, CHRONIC			1 (2%)
PERIVASCULAR CUFFING		1 (2%)	
#URINARY BLADDER	(13)	(33)	(37)
MUCOCELE		1 (3%)	
INFLAMMATION, CHRONIC			1 (3%)
PERIVASCULAR CUFFING		2 (6%)	
ENDOCRINE SYSTEM			
NONE			
REPRODUCTIVE SYSTEM			
#PROSTATE	(19)	(40)	(46)
INFLAMMATION, CHRONIC SUPPURATIV			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1. MALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
NERVOUS SYSTEM			
#BRAIN	(20)	(46)	(50)
INFARCT, NOS			1 (2%)
CALCIFICATION, FOCAL	6 (30%)	7 (15%)	4 (8%)
SPECIAL SENSE ORGANS			
NONE			
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
NONE			
ALL OTHER SYSTEMS			
NONE			
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	3	11	12
ANIMAL MISSING/NO NECROPSY		2	
AUTO/NECROPSY/HISTO PERF	1		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE ADMINISTERED 3(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	20	50	50
ANIMALS MISSING			1
ANIMALS NECROPSIED	20	50	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY	20	50	49
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG/BRONCHUS	(18)	(50)	(49)
CRYSTALS, NOS		1 (2%)	
#LUNG	(18)	(50)	(49)
ATELECTASIS	1 (6%)		
CONGESTION, NOS		1 (2%)	8 (16%)
PNEUMONIA, CHRONIC MURINE	3 (17%)	11 (22%)	3 (6%)
INFLAMMATION, GRANULOMATOUS			1 (2%)
PERIVASCULAR CUFFING			1 (2%)
HISTIOCYTOSIS			1 (2%)
#LUNG/ALVEOLI	(18)	(50)	(49)
CRYSTALS, NOS		1 (2%)	
PHAGOCYTIC CELL		1 (2%)	
HEMATOPOIETIC SYSTEM			
#BONE MARROW	(9)	(46)	(46)
HYPERPLASIA, HEMATOPOIETIC		3 (7%)	1 (2%)
#SPLEEN	(17)	(46)	(45)
HYPERPLASIA, RETICULUM CELL			2 (4%)
HEMATOPOIESIS	4 (24%)	6 (13%)	2 (4%)
#MANDIBULAR L. NODE	(16)	(42)	(40)
INFLAMMATION, GRANULOMATOUS		1 (2%)	
#BRONCHIAL LYMPH NODE	(16)	(42)	(40)
HYPERPLASIA, NOS		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2. FEMALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
#MESENTERIC L. NODE	(16)	(42)	(40)
HEMORRHAGE			1 (3%)
INFLAMMATION, GRANULOMATOUS		1 (2%)	
CYTOMEGALY		1 (2%)	
HYPERPLASIA, RETICULUM CELL		1 (2%)	
HEMATOPOIESIS		1 (2%)	
CIRCULATORY SYSTEM			
#HEART	(18)	(48)	(48)
CALCIFICATION, FOCAL	1 (6%)		
*BLOOD VESSEL	(20)	(50)	(49)
INFLAMMATION, CHRONIC		1 (2%)	
THROMBOPHLEBITIS			1 (2%)
*CORONARY ARTERY	(20)	(50)	(49)
INFLAMMATION, CHRONIC			1 (2%)
*UTERINE VEIN	(20)	(50)	(49)
THROMBOSIS, NOS	1 (5%)		
#HEPATIC SINUSOID	(20)	(50)	(49)
THROMBOSIS, NOS			1 (2%)
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(19)	(48)	(44)
INFLAMMATION, CHRONIC FOCAL		1 (2%)	
PERIVASCULAR CUFFING	2 (11%)	1 (2%)	2 (5%)
#LIVER	(20)	(50)	(49)
INFLAMMATION, NOS		1 (2%)	
INFLAMMATION, CHRONIC FOCAL	2 (10%)	1 (2%)	1 (2%)
INFLAMMATION, GRANULOMATOUS			1 (2%)
PERIVASCULAR CUFFING	1 (5%)	3 (6%)	1 (2%)
DEGENERATION, NOS	1 (5%)		
NECROSIS, FOCAL	1 (5%)	1 (2%)	1 (2%)
INFARCT, NOS		1 (2%)	
METAMORPHOSIS FATTY	1 (5%)		
HYPERPLASIA, NOS	1 (5%)		
ANGIECTASIS			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2. FEMALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
HISTIOCYTOSIS	1 (5%)		
#LIVER/CENTRILOBULAR	(20)	(50)	(49)
NECROSIS, NOS		2 (4%)	
*BILE DUCT	(20)	(50)	(49)
INFLAMMATION, CHRONIC		1 (2%)	1 (2%)
INFLAMMATION, CHRONIC FOCAL		1 (2%)	
#PANCREAS	(13)	(34)	(38)
CYSTIC DUCTS		2 (6%)	
#PANCREATIC ACINUS	(13)	(34)	(38)
INFLAMMATION, CHRONIC		1 (3%)	
ATROPHY, NOS		1 (3%)	
ATROPHY, FOCAL		1 (3%)	1 (3%)
#STOMACH	(19)	(45)	(48)
CYST, NOS			1 (2%)
ABSCESS, NOS			1 (2%)
CALCIFICATION, FOCAL			1 (2%)
HYPERPLASIA, EPITHELIAL			3 (6%)
MASTOCYTOSIS			1 (2%)
#GASTRIC SUBMUCOSA	(19)	(45)	(48)
INFLAMMATION, ACUTE/CHRONIC			1 (2%)
#COLON	(19)	(45)	(47)
PARASITISM	3 (16%)	7 (16%)	4 (9%)
URINARY SYSTEM			
#KIDNEY	(19)	(49)	(49)
CONGESTION, NOS		2 (4%)	9 (18%)
INFLAMMATION, CHRONIC		1 (2%)	1 (2%)
PERIVASCULAR CUFFING	1 (5%)	5 (10%)	4 (8%)
NEPHROSIS, NOS		1 (2%)	
AMYLOIDOSIS		1 (2%)	
METAPLASIA, OSSEOUS			1 (2%)
#KIDNEY/TUBULE	(19)	(49)	(49)
CALCIFICATION, NOS		1 (2%)	
ATROPHY, NOS		1 (2%)	
REGENERATION, NOS		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2. FEMALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
#URINARY BLADDER	(15)	(29)	(36)
INFLAMMATION, CHRONIC	1 (7%)		
PERIVASCULAR CUFFING		2 (7%)	1 (3%)
LYMPHOCYTOSIS			1 (3%)
ENDOCRINE SYSTEM			
#THYROID	(8)	(15)	(36)
HYPERCHROMATISM		1 (7%)	
REPRODUCTIVE SYSTEM			
#UTERUS	(18)	(47)	(47)
CYST, NOS	4 (22%)	1 (2%)	3 (6%)
PYOMETRA		2 (4%)	
#UTERUS/ENDOMETRIUM	(18)	(47)	(47)
CYST, NOS		4 (9%)	1 (2%)
INFLAMMATION, SUPPURATIVE		1 (2%)	1 (2%)
ABSCESS, NOS	1 (6%)		
INFLAMMATION, CHRONIC		1 (2%)	
HYPERPLASIA, CYSTIC	4 (22%)	9 (19%)	6 (13%)
#OVARY/OVIDUCT	(18)	(47)	(47)
INFLAMMATION, NOS		1 (2%)	
HYPERPLASIA, NOS		1 (2%)	
#OVARY	(6)	(13)	(33)
CYST, NOS	1 (17%)	5 (38%)	4 (12%)
HEMORRHAGIC CYST			2 (6%)
NERVOUS SYSTEM			
#BRAIN	(18)	(50)	(49)
PERIVASCULITIS			1 (2%)
CORPORA AMYLACEA	1 (6%)		
CALCIFICATION, NOS	1 (6%)		1 (2%)
CALCIFICATION, FOCAL	2 (11%)	9 (18%)	7 (14%)
#MIDBRAIN	(18)	(50)	(49)
CALCIFICATION, FOCAL			1 (2%)
SPECIAL SENSE ORGANS			
NONE			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2. FEMALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	VEHICLE CONTROL	LOW DOSE	HIGH DOSE
MUSCULOSKELETAL SYSTEM			
*BONE	(20)	(50)	(49)
FIBROUS OSTEODYSTROPHY	7 (35%)	22 (44%)	18 (37%)
OSTEOSCLEROSIS		1 (2%)	
BODY CAVITIES			
*MEDIASTINUM	(20)	(50)	(49)
INFLAMMATION, CHRONIC	1 (5%)		
*PERITONEAL CAVITY	(20)	(50)	(49)
GRANULOMA, NOS		1 (2%)	
*PLEURA	(20)	(50)	(49)
INFLAMMATION, CHRONIC	1 (5%)		
INFLAMMATION, CHRONIC FOCAL			1 (2%)
ALL OTHER SYSTEMS			
NONE			
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	1	2	2
ANIMAL MISSING/NO NECROPSY			1
AUTO/NECROPSY/HISTO PERF			1

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

APPENDIX E

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS IN RATS ADMINISTERED 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Lung: Alveolar/Bronchiolar Carcinoma[b]	2/20 (10)	3/49 (6)	0/49 (0)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		0.612	0.000
Lower Limit		0.078	0.000
Upper Limit		6.996	1.372
Weeks to First Observed Tumor	104	104	--
Hematopoietic System: Leukemia[b]	2/20 (10)	1/50 (2)	2/50 (4)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		0.200	0.400
Lower Limit		0.004	0.032
Upper Limit		3.681	5.277
Weeks to First Observed Tumor	103	104	101

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Stomach: Squamous-cell Papilloma or Carcinoma[b]	0/19 (0)	1/47 (2)	3/50 (6)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		Infinite	Infinite
Lower Limit		0.022	0.238
Upper Limit		Infinite	Infinite
Weeks to First Observed Tumor	--	67	76
Pituitary: Chromophobe Carcinoma[b]	0/19 (0)	3/42 (7)	1/41 (2)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		Infinite	Infinite
Lower Limit		0.284	0.026
Upper Limit		Infinite	Infinite
Weeks to First Observed Tumor	--	89	104

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Pituitary: Chromophobe Adenoma or Carcinoma[b]	1/19 (5)	7/42 (17)	2/41 (5)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		3.167	0.927
Lower Limit		0.460	0.052
Upper Limit		138.815	53.355
Weeks to First Observed Tumor	104	86	104
Adrenal: Pheochromocytoma[b]	2/19 (11)	2/49 (4)	1/49 (2)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		0.388	0.194
Lower Limit		0.031	0.003
Upper Limit		5.108	3.563
Weeks to First Observed Tumor	104	104	101

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Adrenal: Cortical Adenoma or Carcinoma[b]	2/19 (11)	1/49 (2)	1/49 (2)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		0.194	0.194
Lower Limit		0.003	0.003
Upper Limit		3.563	3.563
Weeks to First Observed Tumor	104	104	104
Thyroid: C-cell Adenoma or Carcinoma[b]	1/17 (6)	2/39 (5)	1/41 (2)
P Values[c,d]		N.S.	N.S.
Relative Risk[e]		0.872	0.415
Lower Limit		0.050	0.006
Upper Limit		50.118	31.786
Weeks to First Observed Tumor	104	104	104

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Pancreatic Islets: Islet-cell Adenoma[b]	3/20 (15)	2/48 (4)	0/49 (0)
P Values[c,d]		N.S.	P = 0.022(N)
Relative Risk[e]		0.278	0.000
Lower Limit		0.025	0.000
Upper Limit		2.278	0.673
Weeks to First Observed Tumor	104	104	--
Pancreatic Islets: Islet-cell Adenoma or Carcinoma[b]	3/20 (15)	3/48 (6)	0/49 (0)
P Values[c,d]		N.S.	P = 0.022 (N)
Relative Risk[e]		0.417	0.000
Lower Limit		0.062	0.000
Upper Limit		2.915	0.673
Weeks to First Observed Tumor	104	103	--

Table E1. Anslyses of the Incidence of Primary Tumors in Male Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Testis: Interstitial-cell Tumor[b]	17/19 (89)	38/46 (83)	20/47 (43)
P Values[c,d]		N.S.	P < 0.001(N)
Relative Risk[e]		0.923	0.476
Lower Limit		0.800	0.394
Upper Limit		1.235	0.728
Weeks to First Observed Tumor	104	67	76

[a]Dosed groups received 75 or 150 mg/kg by gavage.

[b]Number of tumor-bearing animals/number of animals examined at site (percent).

[c]Beneath the incidence of tumors in a dosed group is the probability level for the Fisher exact test for the comparison of that dosed group with the control group when P < 0.05; otherwise, not significant (N.S.) is indicated.

[d]A negative trend (N) indicates a lower incidence in a dosed group than in a control group.

[e]The 95% confidence interval of the relative risk between each dosed group and the control group.

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Hematopoietic System: Leukemia[b]	1/20 (5)	3/49 (6)	2/50 (4)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.224	0.800
Lower Limit		0.108	0.045
Upper Limit		62.958	46.273
Weeks to First Observed Tumor	104	76	98
Pituitary: Chromophobe Carcinoma[b]	1/19 (5)	1/43 (2)	1/40 (3)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.442	0.475
Lower Limit		0.006	0.006
Upper Limit		33.913	36.387
Weeks to First Observed Tumor	104	85	95

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Pituitary: Chromophobe Adenoma or Carcinoma[b]	9/19 (47)	12/43 (28)	14/40 (35)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.589	0.739
Lower Limit		0.293	0.385
Upper Limit		1.342	1.619
Weeks to First Observed Tumor	104	85	72
Mammary Gland: Fibroadenoma[b]	2/20 (10)	4/49 (8)	1/50 (2)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.816	0.200
Lower Limit		0.131	0.004
Upper Limit		8.603	3.681
Weeks to First Observed Tumor	104	104	104

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Preputial Gland:			
Adenoma, NOS[b]	2/20 (10)	1/49 (2)	0/50 (0)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.204	0.000
Lower Limit		0.004	0.000
Upper Limit		3.754	1.345
Weeks to First Observed Tumor	104	104	--
Uterus: Endometrial Stromal Polyp[b]	2/19 (11)	8/47 (17)	2/49 (4)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.617	0.388
Lower Limit		0.370	0.031
Upper Limit		14.802	5.108
Weeks to First Observed Tumor	104	76	104

[a]Dosed groups received 75 or 150 mg/kg by gavage.

[b]Number of tumor-bearing animals/number of animals examined at site (percent).

[c]Beneath the incidence of tumors in a dosed group is the probability level for the Fisher exact test for the comparison of that dosed group with the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated.

[d]The 95% confidence interval of the relative risk between each dosed group and the control group.

APPENDIX F

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS IN MICE ADMINISTERED 3-(CHLOROMETHYL)PYRIDINE HYDROCHLORIDE BY GAVAGE

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Lung: Alveolar/Bronchiolar Carcinoma[b]	1/20 (5)	4/45 (9)	5/48 (10)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.778	2.083
Lower Limit		0.195	0.259
Upper Limit		85.520	96.358
Weeks to First Observed Tumor	104	84	102
Lung: Alveolar/Bronchiolar Adenoma or Carcinoma[b]	2/20 (10)	4/45 (9)	5/48 (10)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.889	1.042
Lower Limit		0.143	0.192
Upper Limit		9.340	10.410
Weeks to First Observed Tumor	104	84	102

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Hematopoietic System: Undifferentiated Leukemia[b]	1/20 (5)	3/48 (6)	3/50 (6)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.250	1.200
Lower Limit		0.110	0.106
Upper Limit		64.251	61.724
Weeks to First Observed Tumor	73	94	63
Hematopoietic System: Malignant Lymphoma, Lymphocytic Leukemia or Undifferentiated Leukemia[b]	3/20 (15)	9/48 (19)	9/50 (18)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.250	1.200
Lower Limit		0.361	0.346
Upper Limit		6.662	6.408
Weeks to First Observed Tumor	73	61	63

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Hematopoietic System:			
All Leukemias or Lymphomas[b]	3/20 (15)	9/48 (19)	10/50 (20)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.250	1.333
Lower Limit		0.361	0.398
Upper Limit		6.662	7.002
Weeks to First Observed Tumor	73	61	63
Liver: Hepatocellular Carcinoma[b]	2/20 (10)	5/46 (11)	9/49 (18)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.087	1.837
Lower Limit		0.200	0.434
Upper Limit		10.845	16.572
Weeks to First Observed Tumor	102	58	99

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Liver: Hepatocellular Adenoma or Carcinoma[b]	3/20 (15)	5/46 (11)	9/49 (18)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.725	1.224
Lower Limit		0.160	0.354
Upper Limit		4.348	6.533
Weeks to First Observed Tumor	94	58	99
Stomach: Squamous-cell Papilloma[b]	0/19 (0)	2/43 (5)	8/47 (17)
P Values[c]		N.S.	N.S.
Relative Risk[d]		Infinite	Infinite
Lower Limit		0.136	0.966
Upper Limit		Infinite	Infinite
Weeks to First Observed Tumor	--	61	58

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Stomach: Squamous-cell Papilloma or Carcinoma[b]	0/19 (0)	2/43 (5)	10/47 (21)
P Values[c]		N.S.	P = 0.025
Relative Risk[d]		Infinite	Infinite
Lower Limit		0.136	1.259
Upper Limit		Infinite	Infinite
Weeks to First Observed Tumor	--	61	58

[a]Dosed groups received 100 or 200 mg/kg by gavage.

[b]Number of tumor-bearing animals/number of animals examined at site (percent).

[c]Beneath the incidence of tumors in a dosed group is the probability level for the Fisher exact test for the comparison of that dosed group with the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated.

[d]The 95% confidence interval of the relative risk between each dosed group and the control group.

Table F2. Analyses of the Incidence of Primary Tumors in Female Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Lung: Alveolar/Bronchiolar Carcinoma[b]	1/18 (6)	0/50 (0)	3/49 (6)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.000	1.102
Lower Limit		0.000	0.098
Upper Limit		6.729	56.666
Weeks to First Observed Tumor	65	--	50
Lung: Alveolar/Bronchiolar Adenoma or Carcinoma[b]	1/18 (6)	1/50 (2)	3/49 (6)
P Values[c]		N.S.	N.S.
Relative Risk[d]		0.360	1.102
Lower Limit		0.005	0.098
Upper Limit		27.724	56.666
Weeks to First Observed Tumor	65	104	50

Table F2. Analyses of the Incidence of Primary Tumors in Female Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Hematopoietic System: Malignant Lymphoma, Lymphocytic Leukemia or Undifferentiated Leukemia[b]	2/20 (10)	9/50 (18)	9/49 (18)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.800	1.837
Lower Limit		0.426	0.434
Upper Limit		16.255	16.572
Weeks to First Observed Tumor	93	76	98
Hematopoietic System: All Lymphomas or Leukemias[b]	2/20 (10)	9/50 (18)	10/49 (20)
P Values[c]		N.S.	N.S.
Relative Risk[d]		1.800	2.041
Lower Limit		0.426	0.498
Upper Limit		16.255	18.154
Weeks to First Observed Tumor	93	76	96

Table F2. Analyses of the Incidence of Primary Tumors in Female Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Pituitary: Chromophobe Adenoma[b]	0/9 (0)	0/25 (0)	2/29 (7)
P Values[c]		N.S.	N.S.
Relative Risk[d]		--	Infinite
Lower Limit		--	0.103
Upper Limit		--	Infinite
Weeks to First Observed Tumor	--	--	104
Stomach: Squamous-cell Papilloma[b]	0/19 (0)	1/45 (2)	3/48 (6)
P Values[c]		N.S.	N.S.
Relative Risk[d]		Infinite	Infinite
Lower Limit		0.023	0.248
Upper Limit		Infinite	Infinite
Weeks to First Observed Tumor	--	104	104

Table F2. Analyses of the Incidence of Primary Tumors in Female Mice Administered 3-(Chloromethyl)pyridine Hydrochloride by Gavage[a]

(continued)

Topography: Morphology	Vehicle Control	Low Dose	High Dose
Stomach: Squamous-cell Papilloma or Carcinoma[b]	0/19 (0)	1/45 (2)	5/48 (10)
P Values[c]		N.S.	N.S.
Relative Risk[d]		Infinite	Infinite
Lower Limit		0.023	0.522
Upper Limit		Infinite	Infinite
Weeks to First Observed Tumor	--	104	104

[a]Dosed groups received 100 or 200 mg/kg by gavage.

[b]Number of tumor-bearing animals/number of animals examined at site (percent).

[c]Beneath the incidence of tumors in a dosed group is the probability level for the Fisher exact test for the comparison of that dosed group with the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated.

[d]The 95% confidence interval of the relative risk between each dosed group and the control group.

Review of the Bioassay of 3-(Chloromethyl)pyridine Hydrochloride*
for carcinogenicity
by the Data Evaluation/Risk Assessment Subgroup
of the Clearinghouse on Environmental Carcinogens

June 29, 1978

The Clearinghouse on Environmental Carcinogens was established in May, 1976, in compliance with DHEW Committee Regulations and the Provisions of the Federal Advisory Committee Act. The purpose of the Clearinghouse is to advise the Director of the National Cancer Institute (NCI) on its bioassay program to identify and to evaluate chemical carcinogens in the environment to which humans may be exposed. The members of the Clearinghouse have been drawn from academia, industry, organized labor, public interest groups, State health officials, and quasi-public health and research organizations. Members have been selected on the basis of their experience in carcinogenesis or related fields and, collectively, provide expertise in chemistry, biochemistry, biostatistics, toxicology, pathology, and epidemiology. Representatives of various Governmental agencies participate as ad hoc members. The Data Evaluation/Risk Assessment Subgroup of the Clearinghouse is charged with the responsibility of providing a peer review of reports prepared on NCI-sponsored bioassays of chemicals studied for carcinogenicity. It is in this context that the below critique is given on the bioassay of 3-(Chloromethyl)pyridine Hydrochloride for carcinogenicity.

The reviewer agreed with the conclusion in the report that 3-(Chloromethyl)pyridine Hydrochloride was carcinogenic under the conditions of test. After a brief review of the experimental design, the reviewer said that the study was adequate to support the conclusion on the compound's carcinogenicity. The review of the bioassay of 3-(Chloromethyl) pyridine Hydrochloride was accepted without objection.

Clearinghouse Members present:

Arnold L. Brown (Chairman), Mayo Clinic
Paul Nettesheim, National Institute of Environmental Health Sciences
Verne Ray, Pfizer Medical Research Laboratory
Verald K. Rowe, Dow Chemical U.S.A.
Michael B. Shimkin, University of California at San Diego
Louise Strong, University of Texas Health Sciences Center

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* Subsequent to this review, changes may have been made in the bioassay report either as a result of the review or other reasons. Thus, certain comments and criticisms reflected in the review may no longer be appropriate.

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