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# BIOASSAY OF CARBROMAL FOR POSSIBLE CARCINOGENICITY

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U.S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
Public Health Service
National Institutes of Health

# BIOASSAY OF

# CARBROMAL

# FOR POSSIBLE CARCINOGENICITY

Carcinogenesis Testing Program
Division of Cancer Cause and Prevention
National Cancer Institute
National Institutes of Health
Bethesda, Maryland 20014

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REPORT ON THE BIOASSAY OF CARBROMAL
FOR POSSIBLE CARCINOGENICITY

CARCINOGENESIS TESTING PROGRAM
DIVISION OF CANCER CAUSE AND PREVENTION
NATIONAL CANCER INSTITUTE, NATIONAL INSTITUTES OF HEALTH

FOREWORD: This report presents the results of the bioassay of carbromal 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 chemicals have the capacity to produce cancer in animals. Negative results, in which the test animals do not have a significantly greater incidence of cancer than control animals, do not necessarily mean the test chemical is not a carcinogen because 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 a potential risk to man. The actual determination of the risk to man from animal carcinogens requires a wider analysis.

CONTRIBUTORS: This bioassay of carbromal was conducted by Litton Bionetics, Inc., Kensington, Maryland, initially under direct contract to the NCI and currently under a subcontract to Tracor Jitco, Inc., prime contractor for the NCI Carcinogenesis Testing Program.

The experimental design was determined by the NCI Project Officers, Dr. N. P. Page (1,2), Dr. E. K. Weisburger (1) and Dr. J. H. Weisburger (1,3). The principal investigators for the contract were Dr. F. M. Garner (4) and Dr. B. M. Ulland (4,5). Mr. S. Johnson (4) was the coprincipal investigator for the contract. Animal treatment and observation were supervised by Mr. R. Cypher (4), Mr. D. S. Howard (4) and Mr. H. D. Thornett (4); Mr. H. Paulin (4) analyzed dosed feed mixtures. Ms. J. Blalock (4) was responsible for data collection and assembly. Chemical analysis was performed by Midwest Research Institute (6) and the analytical results were reviewed by Dr. N. Zimmerman (7).

Histopathologic examinations were performed by Dr. P. K. Hildebrandt (4) at Litton Bionetics, Inc., the pathology narratives were written by Dr. P. K Hildebrandt (4), and the diagnoses included in this report represent the interpretation of this pathologist. Histopathology findings and reports were reviewed by Dr. R. L. Schueler (8).

Compilation of individual animal survival, pathology, and summary tables was performed by EG&G Mason Research Institute (9); the statistical analysis was performed by Mr. R. M. Helfand (7) and Dr. J. P. Dirkse, III (10) using methods selected for the Carcinogenesis Testing Program by Dr. J. J. Gart (11).

This report was prepared at METREK, a Division of The MITRE Corporation (7) under the direction of the NCI. Those responsible for this report at METREK are the project coordinator, Dr. L. W. Thomas (7), task leader Ms. P. Walker (7), senior biologist Mr. M. Morse (7), biochemist Mr. S. C. Drill (7), and technical editor Ms. P. A. Miller (7). The final report was reviewed by members of the participating organizations.

The following other scientists at the National Cancer Institute were responsible for evaluating the bioassay experiment, interpreting the results, and reporting the findings: Dr. K. C. Chu (1), Dr. C. Cueto, Jr. (1), Dr. J. F. Douglas (1), Dr. R. A. Griesemer (1), Dr. T. E. Hamm (1), Dr. W. V. Hartwell (1), Dr. M. H. Levitt (1), Dr. H. A. Milman (1), Dr. T. W. Orme (1), Dr. R. A. Squire (1,12), Dr. S. F. Stinson (1), Dr. J. M. Ward (1), and Dr. C. E. Whitmire (1).

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SUMMARY

A bioassay for the possible carcinogenicity of carbromal was conducted using Fischer 344 rats and B6C3F1 mice. Carbromal was administered in the feed, at either of two concentrations, to groups of 50 male and 50 female animals of each species with the exception of 49 low dose male mice and high dose female mice. Twenty animals of each sex and species were placed on test as controls. The high and low dietary concentrations of carbromal were, respectively, 2500 and 1250 ppm for rats and 2500 and 1250 ppm for mice. The compound was administered for 103 weeks to rats and for 78 weeks to mice. The period of compound administration was followed by an observation period of 1 week for rats and 26 weeks for mice.

There were no significant positive associations between the concentrations of carbromal administered and mortality in rats or mice of either sex. Adequate numbers of animals in all groups survived sufficiently long to be at risk from late-developing tumors. Slight dose-related mean body weight depression was observed for male rats and for females of both species and the mean body weight among dosed male mice was lower than that for controls, indicating that the concentrations of carbromal administered to the animals in this bioassay may have approximated the maximum tolerated concentrations.

None of the statistical tests for any site in female rats or in mice of either sex indicated a significant positive association between compound administration and tumor incidence. There was a significant positive association between the concentrations administered and the incidences of adrenal pheochromocytomas in male rats; however, the Fisher exact comparisons were not significant.

Under the conditions of this bioassay, dietary administration of carbromal was not carcinogenic in Fischer 344 rats or B6C3F1 mice.

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# I. INTRODUCTION

Carbromal (Figure 1) (NCI No. CO3805), a mild central nervous system depressant, was selected for bioassay by the National Cancer Institute because of the similarity of the biological activity of this compound to that of urethan, which is known to induce leukemia in mice (Vesselinovitch, 1968) and is an initiator of skin carcinogenesis in mice (Roe and Salaman, 1955), and the widespread exposure to this compound among the general population via deliberate ingestion for medicinal purposes.

The Chemical Abstracts Service (CAS) Ninth Collective Index (1977) name for this compound is N-(aminocarbonyl)-2-bromo-2-ethylbutanamide.* It is also known as (2-bromo-2-ethylbutyryl)urea, bromodiethylacetylcarbamide, bromodiethylacetylurea and ($\alpha$-bromo-$\alpha$-ethylbutyryl)carbamide.

Carbromal is a mild sedative and hypnotic drug, similar in its biological action to both urethan (Roe and Salaman, 1955) and the barbiturates (Gosselin et al., 1976). The usual prescribed dosage is 0.3 to 0.6 grams (Gosselin et al., 1976). Carbromal is at least partially decomposed to release the bromide ion in the body, but the sedative action of the compound is presumably due to the intact molecule (Gosselin et al., 1976).

Specific production data for carbromal are not available; however, one U.S. company currently manufactures carbromal in commercial

*The CAS registry number is 77-65-6.

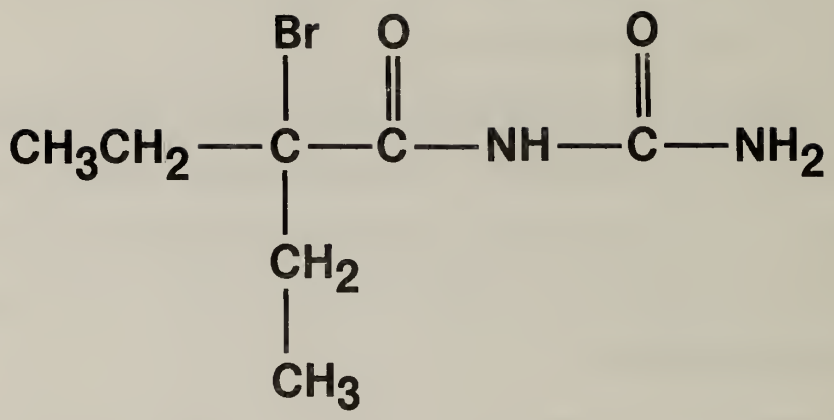



FIGURE 1
CHEMICAL STRUCTURE OF CARBROMAL

quantities (in excess of 1000 pounds or $1000 in value annually) (Stanford Research Institute, 1977).

Acute carbromal poisoning leads to progressive depression of the central nervous system, manifested as drowsiness, stupor, coma, and death due to respiratory failure (Gosselin et al., 1976). Recovery is probable after ingestion of as much as 3 grams (Gosselin et al., 1976).

## II. MATERIALS AND METHODS

### A. Chemicals

Carbromal was purchased from Pfaltz & Bauer, Inc., Stamford, Connecticut. Chemical analysis was performed by Midwest Research Institute, Kansas City, Missouri. The experimentally determined melting point of 118° to 119°C compared favorably to that found in the literature (117° to 118°C) (Rosenmund and Herrmann, 1912). The results of infrared (IR) and nuclear magnetic resonance (NMR) analyses were consistent with those found in the literature (Sadtler Standard Spectra). Thin-layer chromatography (TLC) was performed utilizing two solvent systems (i.e., benzene:acetone and ethyl acetate). Each plate was visualized with 254 and 367 nm light, dichromate, and heat. In each case one major spot and one less motile impurity were observed. Elemental analysis was consistent with that expected based on the molecular formula of the compound, $C_7H_{13}BrN_2O_2$. One homogeneous peak was observed using high pressure liquid chromatography (HPLC).

A second batch of the compound was received from the same company approximately one year later. The experimentally determined melting point of this batch was 116.5° to 119°C. The results of IR and NMR analyses were consistent with those found in the literature. TLC, performed utilizing the same solvent systems and method of visualization used previously, revealed one major spot on each plate.

The results of elemental analysis and HPLC were consistent with those reported for the first batch.

Throughout this report, the term carbromal is used to represent this material.

B. Dietary Preparation

The basal laboratory diet for both dosed and control animals consisted of Wayne Lab-Blox® meal (Allied Mills, Inc., Chicago, Illinois). Carbromal was administered to the dosed animals as a component of the diet.

The chemical was removed from its container and a proper amount was blended with an aliquot of the feed using a mortar and pestle. Once visual homogeneity was attained, the mixture was placed in a 6 kg capacity Patterson-Kelley standard model twin-shell stainless steel V-blender along with the remainder of the feed to be prepared. After 20 minutes of blending, the mixtures were placed in double plastic bags and stored in the dark at 4°C. The mixture was prepared once weekly.

Dosed feed preparations containing 1250 and 2500 ppm of carbromal were analyzed spectrophotometrically. The mean result immediately after preparation was 100 percent of theoretical (ranging from 96 to 104 percent). After 10 days at ambient room temperature, the mean result was 94 percent of theoretical.

C. Animals

The two animal species, Fischer 344 rats and B6C3F1 mice, used in the carcinogenicity bioassay were obtained through contracts of the Division of Cancer Treatment, National Cancer Institute. All rats were supplied by A. R. Schmidt, Madison, Wisconsin. All mice were supplied by Charles River Breeding Laboratories, Inc., Wilmington, Massachusetts.

Rats and mice were approximately 4 weeks old when received. Upon receipt, animals were examined and any obviously ill or runted animals were killed. The remaining animals were quarantined for 2 weeks prior to initiation of test. Animals which did not manifest clinical signs of disease were placed on test at this time. Animals were assigned to groups and distributed among cages so that the average body weight per cage was approximately equal for a given species and sex.

D. Animal Maintenance

All animals were housed by species in rooms with a temperature range of 22° to 26°C and a range in relative humidity of 45 to 55 percent. Incoming air was filtered through HEPA filters (Flanders Filters, McLean, Virginia) at a rate of 12 to 15 complete changes of room air per hour. Fluorescent lighting was provided 8 hours per day (9:00 a.m. to 5:00 p.m.).

All rats were housed four per cage by sex and all mice were housed five per cage by sex. Throughout the study dosed and control

animals of both species were housed in polycarbonate cages (Lab Products, Inc., Garfield, New Jersey) suspended from aluminum racks. Racks were fitted with a continuous piece of stainless steel mesh over which a sheet of filter paper was firmly secured. Filter paper was changed at 2-week intervals, when the racks were sanitized. Clean cages and bedding were provided twice weekly. Ab-sorb-dri® hardwood chip bedding (Wilner Wood Products Company, Norway, Maine) was used in polycarbonate cages for the entire bioassay.

Acidulated water (pH 2.5) was supplied to animals in water bottles. Water bottles were changed and washed twice weekly, and sipper tubes were washed at weekly intervals. During the period of chemical administration, dosed and control animals received treated or untreated Wayne Lab-Blox® meal as appropriate. The feed was supplied in hanging stainless steel hoppers which were refilled three times per week and sanitized weekly. Food and water were available *ad libitum* for both species.

All dosed and control rats were housed in a room with other rats receiving diets containing* triphenyltin hydroxide (76-87-9) and diaminozide (1596-84-5); and other rats intubated with β-nitrostyrene (102-96-5).

All dosed and control mice were housed in a room with mice receiving diets containing EDTA trisodium salt (150-38-9); 3,3'-dimethoxybenzidine-4,4'-diisocyanate (91-93-0); triphenyltin hydroxide

*CAS registry numbers are given in parentheses.

(76-87-9); N,N'-diethylthiourea (105-55-5); diaminozide (1596-84-5); p-quinone dioxime (105-11-3); 4-amino-2-nitrophenol (119-34-6); other mice intubated with lithocholic acid (434-13-9); and with other mice receiving I.P. injections of methiodal sodium (126-31-8).

E. Selection of Initial Concentrations

To establish the concentrations of carbromal for administration to dosed animals in the chronic studies, subchronic toxicity tests were conducted with both rats and mice. Animals of each species were distributed among nine groups, each consisting of five males and five females. Carbromal was incorporated into the basal laboratory diet and supplied ad libitum to seven of the nine groups of each species in concentrations of 1470, 2160, 3150, 4600, 6800, 10,000 and 14,700 ppm. The two remaining groups of each species served as control groups, receiving only the basal laboratory diet.

The dosed dietary preparations were administered for a period of 7 weeks, followed by a 1-week observation period during which all animals were fed the basal laboratory diet. Individual body weights and food consumption data were recorded twice weekly throughout the study. Upon termination of the study all survivors were sacrificed and necropsied.

The following table indicates the mean body weight gain, relative to controls, survival and incidence of abnormal clinical signs observed in each of the dosed rat groups at the end of the subchronic test.

RAT SUBCHRONIC STUDY RESULTS

| ppm | Mean Body Weight Gain (%)[a] | | Survival[b] | | Observation of Abnormal Clinical Signs[b] | |
|---|---|---|---|---|---|---|
| | Males | Females | Males | Females | Males | Females |
| 14,700 | -47 | -18 | 5/5 | 5/5 | 5/5[c,d] | 5/5[c,d] |
| 10,000 | -46 | -14 | 5/5 | 5/5 | 5/5[c,d] | 5/5[c,d] |
| 6,800 | -36 | - 5 | 5/5 | 5/5 | 5/5[d] | 5/5[d] |
| 4,600 | -40 | + 2 | 5/5 | 5/5 | 5/5[d] | 5/5[d] |
| 3,150 | + 1 | 0 | 5/5 | 5/5 | 5/5[d] | 5/5[d] |
| 2,160 | -16 | 0 | 5/5 | 5/5 | 0/5 | 0/5 |
| 1,470 | - 4 | + 9 | 5/5 | 5/5 | 0/5 | 0/5 |
| 0 | -- | -- | 5/5 | 5/5 | 0/5 | 0/5 |

The high concentration selected for administration to dosed rats in the chronic bioassay was 2500 ppm.

The following table indicates the mean body weight gain, relative to controls, and survival observed in each of the dosed mouse groups at the end of the subchronic test.

MOUSE SUBCHRONIC STUDY RESULTS

| ppm | Mean Body Weight Gain (%)[a] | | Survival[b] | |
|---|---|---|---|---|
| | Males | Females | Males | Females |
| 14,700 | -26 | -14 | 5/5 | 2/5 |
| 10,000 | -14 | -11 | 5/5 | 2/5 |
| 6,800 | - 4 | - 8 | 5/5 | 5/5 |
| 4,600 | 0 | - 4 | 5/5 | 4/5 |
| 3,150 | - 7 | + 7 | 5/5 | 5/5 |
| 2,160 | +11 | - 2 | 5/5 | 5/5 |
| 1,470 | + 2 | + 2 | 5/5 | 5/5 |
| 0 | -- | -- | 5/5 | 5/5 |

[a] + is indicative of mean body weight gain greater than that of controls
- is indicative of mean body weight gain less than that of controls.
[b] Number of animals observed/number of animals originally in group.
[c] These rats exhibited rough coats and lack of coordination.
[d] These rats had mottled livers.

No abnormal clinical signs were recorded. The high concentration selected for administration to dosed mice in the chronic bioassay was 2500 ppm.

F. Experimental Design

The experimental design parameters for the chronic study (species, sex, group size, concentrations administered, and duration of treated and untreated observation periods) are summarized in Tables 1 and 2.

All rats were approximately 6 weeks old at the time the test was initiated and were placed on test simultaneously. The dietary concentrations of carbromal administered to rats were 2500 and 1250 ppm. Throughout this report those rats receiving the former concentration are referred to as the high dose groups and those receiving the latter concentration are referred to as the low dose groups. Dosed rats were supplied with feed containing carbromal for 103 weeks followed by a 1-week observation period.

All mice were approximately 6 weeks old at the time the test was initiated and were placed on test simultaneously. The dietary concentrations of carbromal administered were 2500 and 1250 ppm. Throughout this report those mice receiving the former concentration are referred to as the high dose groups and those receiving the latter concentration are referred to as the low dose groups. Dosed mice were supplied with feed containing carbromal for 78 weeks followed by a 26-week observation period.

TABLE 1

DESIGN SUMMARY FOR FISCHER 344 RATS
CARBROMAL FEEDING EXPERIMENT

| | INITIAL GROUP SIZE | CARBROMAL CONCENTRATION[a] | OBSERVATION PERIOD | |
|---|---|---|---|---|
| | | | TREATED (WEEKS) | UNTREATED (WEEKS) |
| MALE | | | | |
| CONTROL | 20 | 0 | 0 | 104 |
| LOW DOSE | 50 | 1250 | 103 | |
| | | 0 | | 1 |
| HIGH DOSE | 50 | 2500 | 103 | |
| | | 0 | | 1 |
| FEMALE | | | | |
| CONTROL | 20 | 0 | 0 | 104 |
| LOW DOSE | 50 | 1250 | 103 | |
| | | 0 | | 1 |
| HIGH DOSE | 50 | 2500 | 103 | |
| | | 0 | | 1 |

[a]Concentrations given in parts per million.

TABLE 2

DESIGN SUMMARY FOR B6C3F1 MICE
CARBROMAL FEEDING EXPERIMENT

| | INITIAL GROUP SIZE | CARBROMAL CONCENTRATION[a] | OBSERVATION PERIOD TREATED (WEEKS) | OBSERVATION PERIOD UNTREATED (WEEKS) |
|---|---|---|---|---|
| MALE | | | | |
| CONTROL | 20 | 0 | 0 | 104 |
| LOW DOSE | 49 | 1250 | 78 | |
| | | 0 | | 26 |
| HIGH DOSE | 50 | 2500 | 78 | |
| | | 0 | | 26 |
| FEMALE | | | | |
| CONTROL | 20 | 0 | 0 | 104 |
| LOW DOSE | 50 | 1250 | 78 | |
| | | 0 | | 26 |
| HIGH DOSE | 49 | 2500 | 78 | |
| | | 0 | | 26 |

[a]Concentrations given in parts per million.

G. Clinical and Histopathologic Examinations

Animals were weighed immediately prior to initiation of the experiment. Body weights of rats were recorded once a week for the first 3 weeks and at monthly intervals thereafter. Body weights of mice were recorded once a week for the first 6 weeks, once every 2 weeks for the next 6 weeks, and at monthly intervals thereafter. All animals were inspected twice daily. Food consumption data were collected at monthly intervals from 20 percent of the animals in each group.

All moribund animals or animals that developed large, palpable masses that jeopardized their health were sacrificed. A necropsy was performed on each animal regardless of whether it died, was sacrificed when moribund, or was sacrificed at the end of the bioassay. The animals were euthanized by carbon dioxide asphyxiation, and were immediately necropsied. The histopathologic examination consisted of gross and microscopic examination of all major tissues, organs, and gross lesions taken from sacrificed animals and, whenever possible, from animals found dead.

Tissues were preserved in a 10 percent neutral buffered formalin solution, embedded in paraffin, sectioned, and stained with hematoxylin and eosin prior to microscopic examination.

Slides were prepared from the following tissues: skin, subcutaneous tissue, lungs and bronchi, trachea, bone marrow, spleen, lymph nodes, thymus, heart, salivary gland, liver, gallbladder (mice),

pancreas, esophagus, stomach, small intestine, large intestine, kidney, urinary bladder, pituitary, adrenal, thyroid, parathyroid, testis, prostate, brain, uterus, mammary gland, and ovary.

A few tissues were not examined for some animals, particularly for those that died early. Also, some animals were missing, cannibalized, or judged to be in such an advanced state of autolysis as to preclude histopathologic interpretation. Thus, the number of animals for which particular organs, tissues, or lesions were examined microscopically varies and does not necessarily represent the number of animals that were recorded in each group at the time that the test was initiated.

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) when testing two groups for equality and used Tarone's (1975) extensions of Cox's methods when testing 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 was 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, pp. 48-52) was used to compare the tumor incidence of a control group to that of a group of treated animals at each dose level. When results for a number of treated 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, pp. 6-10) 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, pp. 362-365), was also used when appropriate. Under the assumption of a linear trend, this test determined 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 was 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 animals died naturally or were 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 treated 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 treated 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 treated 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 percent of a large number of identical experiments, the true ratio of the risk in a treated 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 (a $P < 0.025$ 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. CHRONIC TESTING RESULTS: RATS

### A. Body Weights and Clinical Observations

Distinct and consistent dose-related mean body weight depression was apparent in both male and female rats (Figure 2).

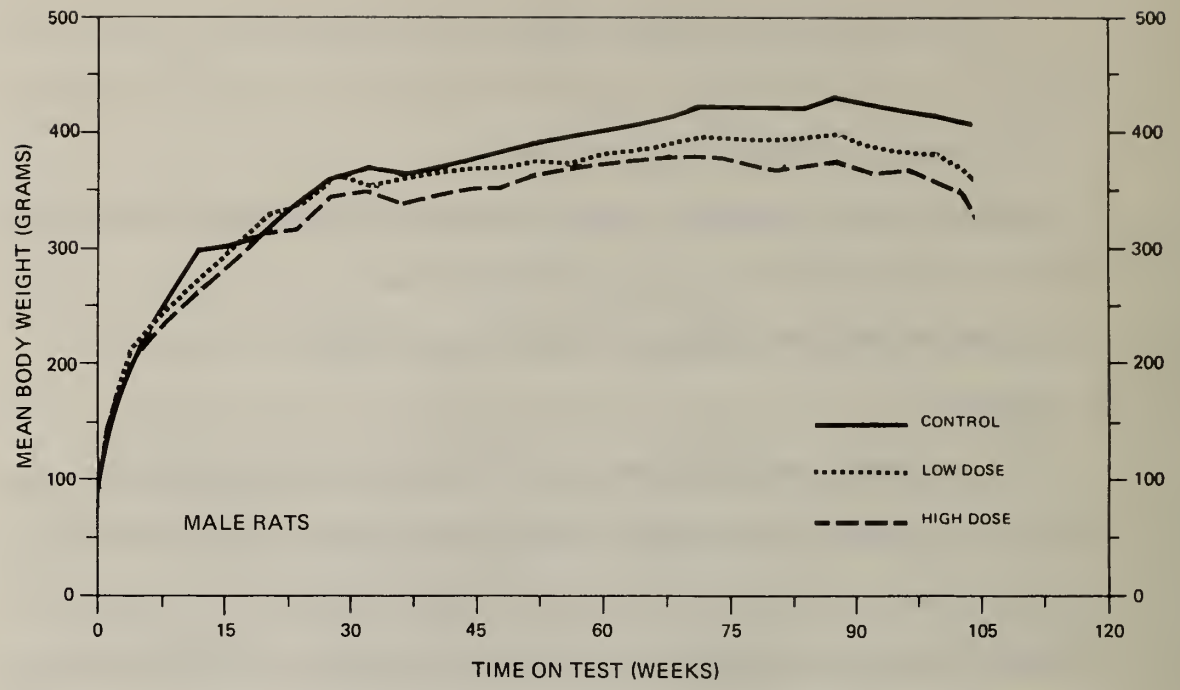
No other abnormal clinical signs were recorded.

### B. Survival

The estimated probabilities of survival for male and female rats in the control and carbromal-dosed groups are shown in Figure 3. The Tarone test did not indicate a significant positive association between dosage and mortality for rats of either sex. The individual Cox tests also indicated no significant positive associations for male or female rats. The Tarone test for male rats, however, did indicate a significant negative association, as did the Cox test comparing the high dose group to the control group.

There were adequate numbers of male rats at risk from late-developing tumors as 90 percent (45/50) of the high dose, 86 percent (43/50) of the low dose, and 65 percent (13/20) of the controls survived on test for at least 104 weeks.

There were also adequate numbers of female rats at risk from late-developing tumors, as 90 percent (45/50) of the high dose, 82 percent (41/50) of the low dose, and 85 percent (17/20) of the controls survived on test until the termination of the study.



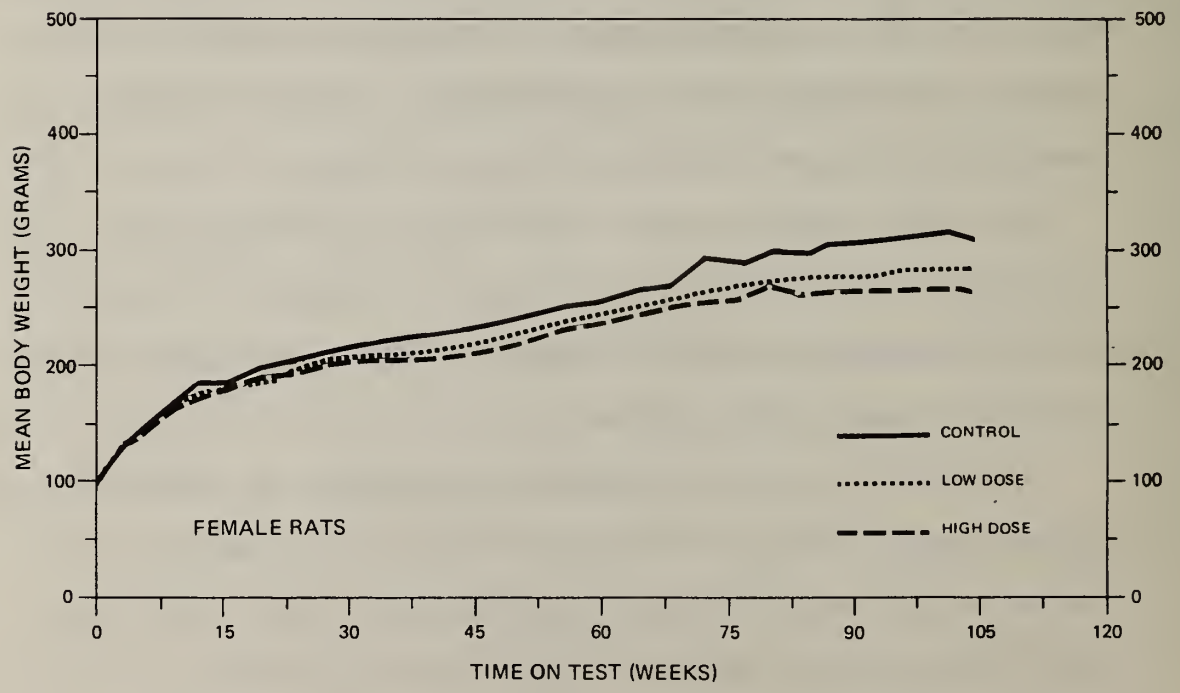
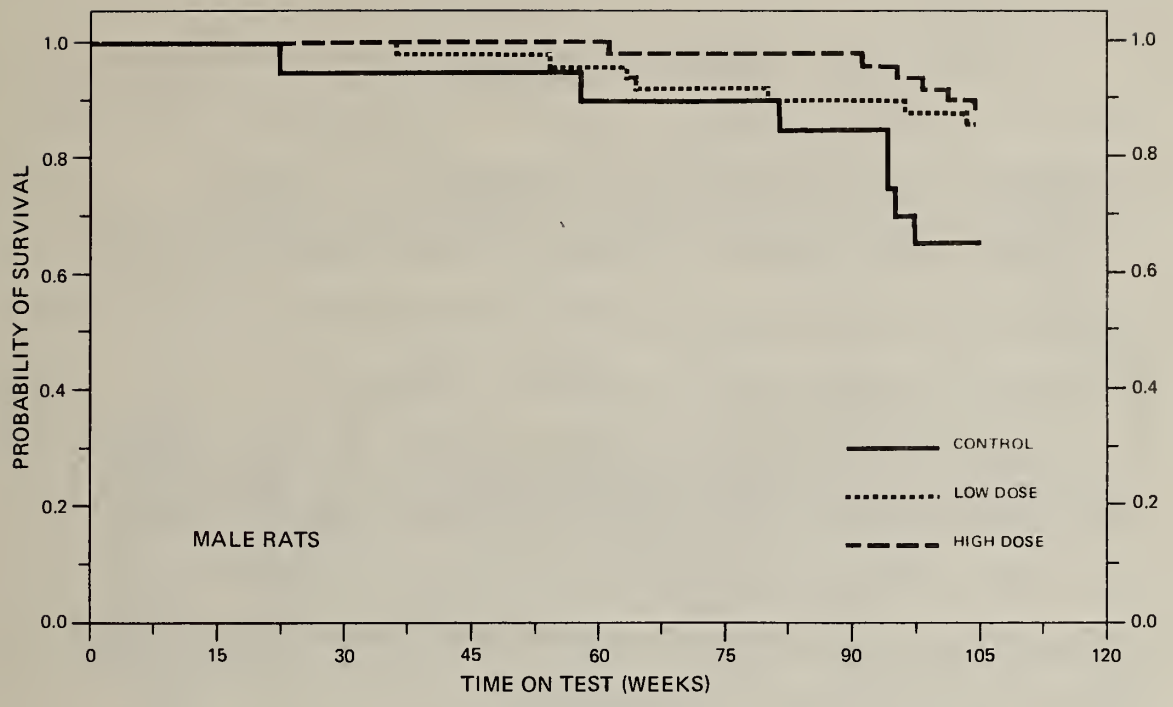




**FIGURE 2**
**GROWTH CURVES FOR CARBROMAL CHRONIC RATS**



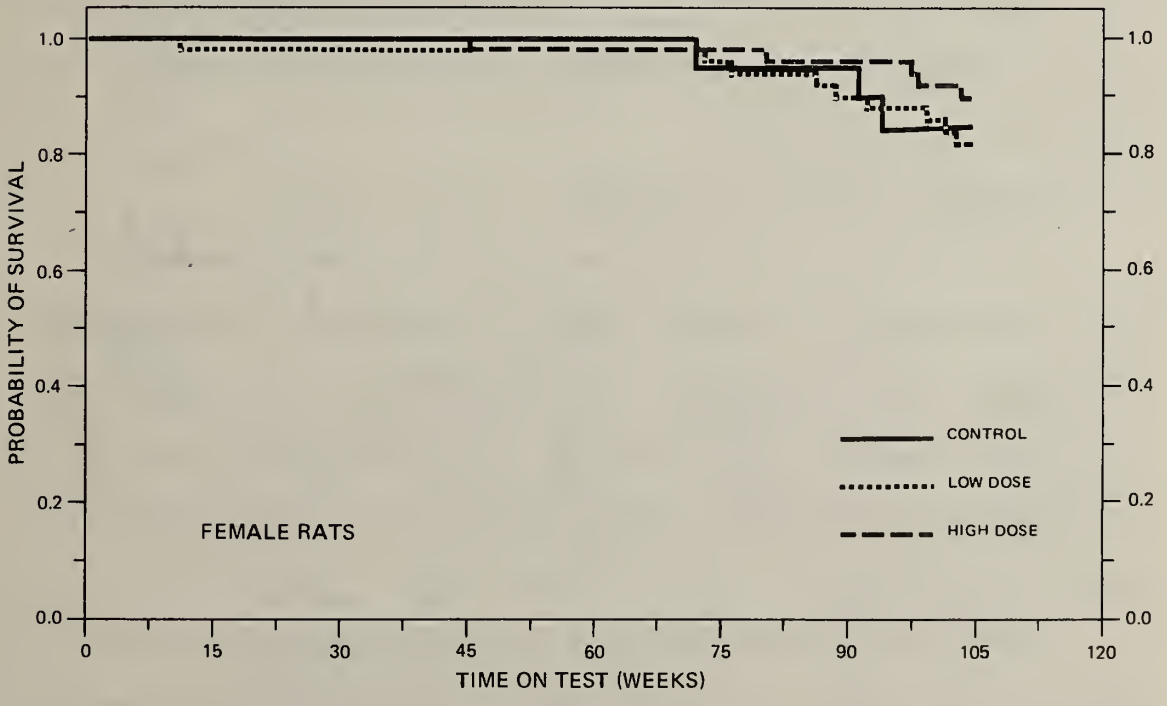




**FIGURE 3**
**SURVIVAL COMPARISONS OF CARBROMAL CHRONIC STUDY RATS**

C. Pathology

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).

There was an increased incidence of pheochromocytoma of the adrenal in the male high dose group (1/19 [5 percent] controls, 2/49 [4 percent] low dose, 8/46 [17 percent] high dose).

With the exception of the lesion referred to above, the pathologic changes observed occurred in nearly equal numbers among control and dosed animals, and for each sex appeared to be within the range commonly observed in aging Fischer 344 rats. These lesions were, therefore, considered to be spontaneous.

Based upon the results of this pathologic examination, carbromal was not considered to be carcinogenic to Fischer 344 rats under the conditions of this bioassay.

D. Statistical Analyses of Results

The results of the statistical analyses of tumor incidence in rats are summarized in Tables 3 and 4. The analysis is included for every type of malignant tumor in either sex where at least two such tumors were observed in at least one of the control or carbromal-dosed groups and where such tumors were observed in at least 5 percent of the group.

The Cochran-Armitage test indicated a significant ($P = 0.043$) positive association between dose and the incidence of pheochromocytomas of the adrenal in male rats. However, the Fisher exact tests

TABLE 3

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN MALE RATS TREATED WITH CARBROMAL[a]

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Hematopoietic System: Leukemia or Malignant Lymphoma[b] | 3/20(0.15) | 5/50(0.10) | 3/50(0.06) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.667 | 0.400 |
| Lower Limit | --- | 0.147 | 0.060 |
| Upper Limit | --- | 4.014 | 2.802 |
| Weeks to First Observed Tumor | 81 | 64 | 61 |
| Pituitary: Chromophobe Adenoma[b] | 0/17(0.00) | 2/41(0.05) | 3/40(0.07) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | Infinite | Infinite |
| Lower Limit | --- | 0.129 | 0.269 |
| Upper Limit | --- | Infinite | Infinite |
| Weeks to First Observed Tumor | --- | 104 | 104 |
| Adrenal: Pheochromocytoma[b] | 1/19(0.05) | 2/49(0.04) | 8/46(0.17) |
| P Values[c] | P = 0.043 | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.776 | 3.304 |
| Lower Limit | --- | 0.044 | 0.501 |
| Upper Limit | --- | 44.838 | 142.909 |
| Weeks to First Observed Tumor | 95 | 103 | 61 |

TABLE 3 (CONTINUED)

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Pancreatic Islets: Islet-Cell Carcinoma or Islet-Cell Adenoma[b] | 0/18(0.00) | 0/45(0.00) | 3/44(0.07) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | --- | Infinite |
| Lower Limit | --- | --- | 0.258 |
| Upper Limit | --- | --- | Infinite |
| Weeks to First Observed Tumor | --- | --- | 104 |
| Testis: Interstitial-Cell Tumor[b] | 18/20(0.90) | 45/49(0.92) | 47/49(0.96) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 1.020 | 1.066 |
| Lower Limit | --- | 0.895 | 0.937 |
| Upper Limit | --- | 1.256 | 1.220 |
| Weeks to First Observed Tumor | 81 | 96 | 91 |
| Body Cavities: Mesothelioma NOS[b] | 1/20(0.05) | 3/50(0.06) | 0/50(0.00) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 1.200 | 0.000 |
| Lower Limit | --- | 0.106 | 0.000 |
| Upper Limit | --- | 61.724 | 7.475 |
| Weeks to First Observed Tumor | 104 | 96 | --- |

TABLE 3 (CONCLUDED)

[a]Treated groups received doses of 1250 or 2500 ppm in feed.

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

[c]The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d]The 95% confidence interval on the relative risk of the treated group to the control group.

TABLE 4

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT
SPECIFIC SITES IN FEMALE RATS TREATED WITH CARBROMAL[a]

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Hematopoietic System: Leukemia or Malignant Lymphoma[b] | 3/20(0.15) | 4/50(0.08) | 4/50(0.08) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.533 | 0.533 |
| Lower Limit | --- | 0.102 | 0.102 |
| Upper Limit | --- | 3.410 | 3.410 |
| Weeks to First Observed Tumor | 91 | 76 | 80 |
| Pituitary: Chromophobe Adenoma or Acidophil Adenoma[b] | 4/19(0.21) | 11/46(0.24) | 16/49(0.33) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 1.136 | 1.551 |
| Lower Limit | --- | 0.400 | 0.599 |
| Upper Limit | --- | 4.428 | 5.745 |
| Weeks to First Observed Tumor | 104 | 88 | 97 |
| Thyroid: C-Cell Adenoma[b] | 1/17(0.06) | 1/36(0.03) | 2/35(0.06) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.472 | 0.971 |
| Lower Limit | --- | 0.006 | 0.056 |
| Upper Limit | --- | 36.073 | 55.675 |
| Weeks to First Observed Tumor | 104 | 104 | 104 |

TABLE 4 (CONCLUDED)

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Mammary Gland: Fibroadenoma[b] | 3/20(0.15) | 3/50(0.06) | 2/50(0.04) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.400 | 0.267 |
| Lower Limit | --- | 0.060 | 0.024 |
| Upper Limit | --- | 2.802 | 2.190 |
| Weeks to First Observed Tumor | 104 | 88 | 104 |
| Uterus: Endometrial Stromal Polyp[b] | 2/20(0.10) | 2/47(0.04) | 4/47(0.09) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.426 | 0.851 |
| Lower Limit | --- | 0.034 | 0.136 |
| Upper Limit | --- | 5.603 | 8.956 |
| Weeks to First Observed Tumor | 104 | 104 | 104 |

[a]Treated groups received doses of 1250 or 2500 ppm in feed.

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

[c]The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d]The 95% confidence interval on the relative risk of the treated group to the control group.

comparing high dose to control and low dose to control were not significant.

Neither the Cochran-Armitage nor the Fisher exact tests were significant at any site in female rats.

To provide additional insight into the possible carcinogenicity of this compound, 95 percent confidence intervals on the relative risk have been estimated and entered in the tables based upon the observed tumor incidence rates. In many of the intervals shown in Tables 3 and 4, the value one is included; this indicates the absence of statistically significant results. It should also be noted that many of the confidence intervals have an upper limit greater than one, indicating the theoretical possibility of tumor induction in rats by carbromal that could not be established under the conditions of this test.

## IV. CHRONIC TESTING RESULTS: MICE

### A. Body Weights and Clinical Observations

Dosed male mice evidenced mean body weight depression relative to controls. Distinct and consistent dose-related mean body weight depression was observed in female mice throughout the bioassay (Figure 4).

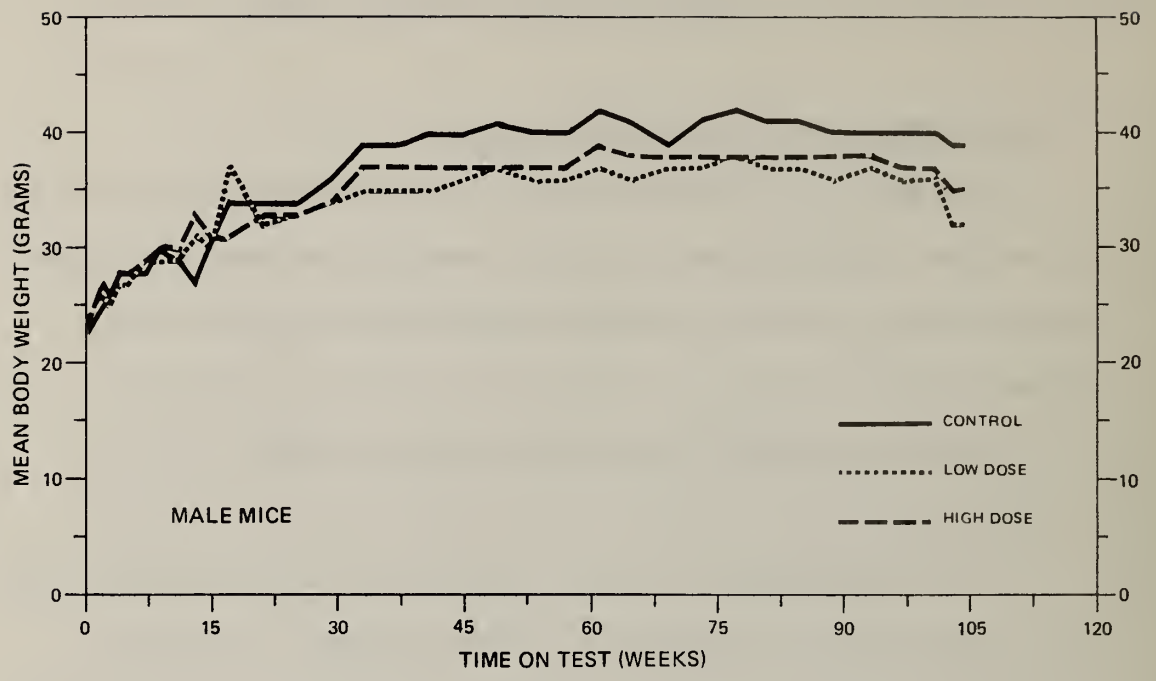
No other abnormal clinical signs were recorded.

### B. Survival

The estimated probabilities of survival for male and female mice in the control and carbromal-dosed groups are shown in Figure 5. Neither the Tarone test for association between dosage and mortality nor the individual Cox tests were significant for either male or female mice.

There were adequate numbers of male mice at risk from late-developing tumors, as 84 percent (42/50) of the high dose, 61 percent (30/49) of the low dose and 90 percent (18/20) of the controls survived on test for at least 104 weeks. Fourteen low dose male mice were reported as missing: 5 in week 16, 5 in week 19, and 4 in week 33.

There were also an adequate number of female mice at risk from late-developing tumors, as 84 percent (41/49) of the high dose, 72 percent (36/50) of the low dose and 75 percent (15/20) of the controls survived on test until the termination of the study.



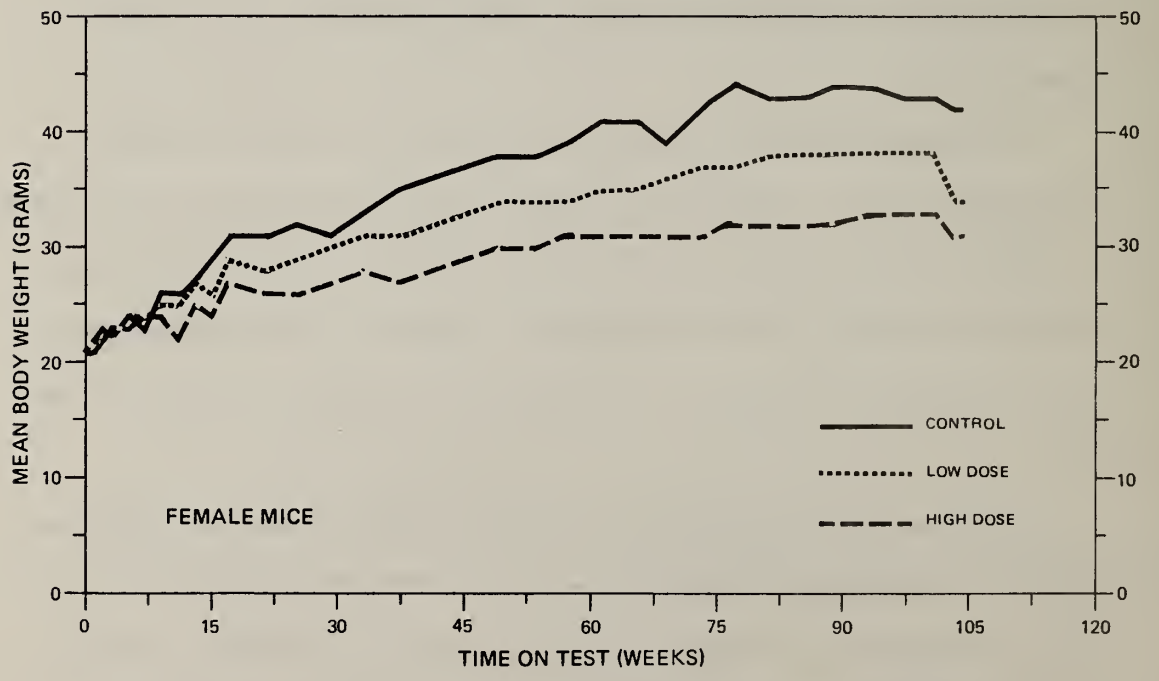
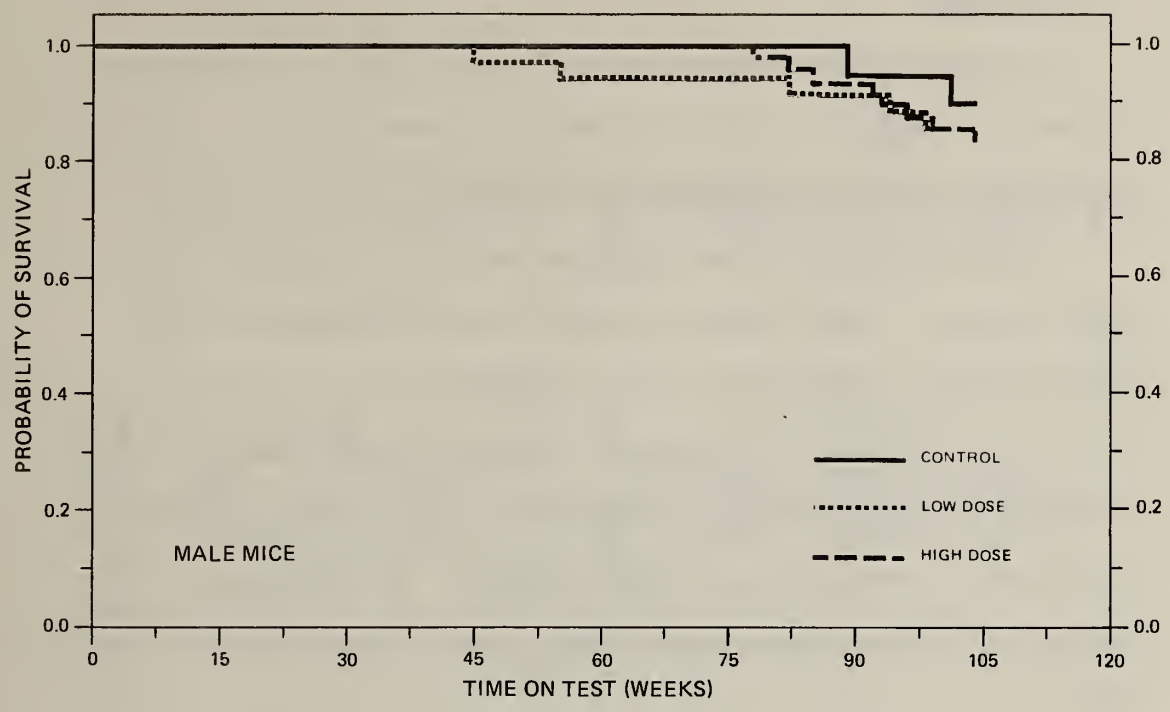




**FIGURE 4**
**GROWTH CURVES FOR CARBROMAL CHRONIC STUDY MICE**



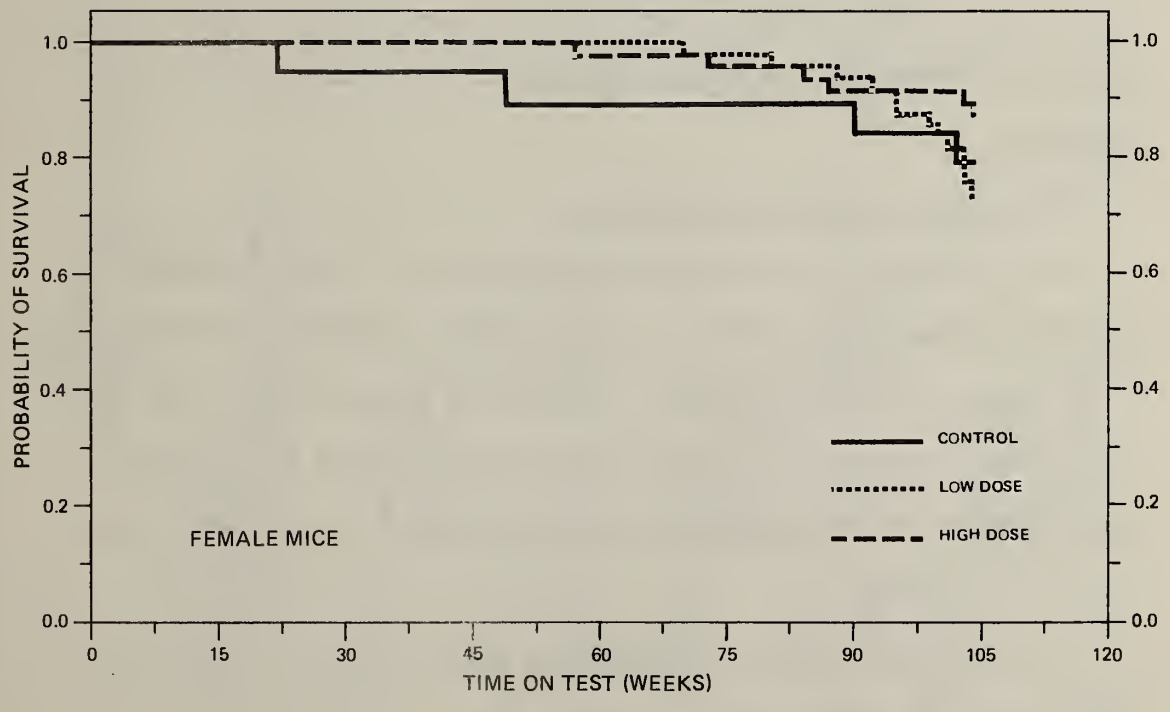




**FIGURE 5**
**SURVIVAL COMPARISONS OF CARBROMAL CHRONIC STUDY MICE**

C. Pathology

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 tumors was observed both in the control group and dosed groups. These lesions, however, are not uncommon in this strain of mice independent of any treatment.

In addition to the neoplastic lesions, a large number of degenerative, proliferative, and inflammatory changes were encountered in animals of the dosed and control groups. No differences in the incidence of these lesions were found between dosed mice and control mice.

The results of this pathologic study indicated that carbromal was not carcinogenic in B6C3F1 mice under the conditions of this bioassay.

D. Statistical Analyses of Results

The results of the statistical analyses of tumor incidence in mice are summarized in Tables 5 and 6. The analysis is included for every type of malignant tumor in either sex where at least two such tumors were observed in at least one of the control or carbromal-dosed groups and where such tumors were observed in at least 5 percent of the group.

The Cochran-Armitage test was not significant at any site in either male or female mice.

TABLE 5

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN MALE MICE TREATED WITH CARBROMAL[a]

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Lung: Alveolar/Bronchiolar Carcinoma or Alveolar/Bronchiolar Adenoma[b] | 7/20(0.35) | 4/34(0.12) | 8/46(0.17) |
| P Values[c] | N.S. | P = 0.046(N) | N.S. |
| Relative Risk (Control)[d] | --- | 0.336 | 0.497 |
| Lower Limit | --- | 0.085 | 0.191 |
| Upper Limit | --- | 1.162 | 1.419 |
| Weeks to First Observed Tumor | 101 | 104 | 82 |
| Hematopoietic System: Leukemia or Malignant Lymphoma[b] | 3/20(0.15) | 3/35(0.09) | 5/49(0.10) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 0.571 | 0.680 |
| Lower Limit | --- | 0.086 | 0.150 |
| Upper Limit | --- | 3.941 | 4.092 |
| Weeks to First Observed Tumor | 89 | 94 | 93 |
| Liver: Hepatocellular Carcinoma[b] | 3/20(0.15) | 6/35(0.17) | 6/48(0.13) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 1.143 | 0.833 |
| Lower Limit | --- | 0.281 | 0.204 |
| Upper Limit | --- | 6.470 | 4.799 |
| Weeks to First Observed Tumor | 101 | 82 | 104 |

TABLE 5 (CONCLUDED)

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Liver: Hepatocellular Carcinoma or Hepatocellular Adenoma[b] | 4/20(0.20) | 8/35(0.23) | 13/48(0.27) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 1.143 | 1.354 |
| Lower Limit | --- | 0.360 | 0.495 |
| Upper Limit | --- | 4.644 | 5.170 |
| Weeks to First Observed Tumor | 101 | 82 | 85 |

[a]Treated groups received doses of 1250 or 2500 ppm in feed.

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

[c]The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d]The 95% confidence interval on the relative risk of the treated group to the control group.

TABLE 6

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN FEMALE MICE TREATED WITH CARBROMAL[a]

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Lung: Alveolar/Bronchiolar Carcinoma or Alveolar/Bronchiolar Adenoma[b] | 0/18(0.00) | 8/49(0.16) | 9/47(0.19) |
| P Values[c] | N.S. | N.S. | P = 0.043 |
| Relative Risk (Control)[d] | --- | Infinite | Infinite |
| Lower Limit | --- | 0.881 | 1.058 |
| Upper Limit | --- | Infinite | Infinite |
| Weeks to First Observed Tumor | --- | 99 | 103 |
| Hematopoietic System: Leukemia or Malignant Lymphoma[b] | 6/19(0.32) | 17/49(0.35) | 13/47(0.28) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | 1.099 | 0.876 |
| Lower Limit | --- | 0.509 | 0.381 |
| Upper Limit | --- | 2.974 | 2.469 |
| Weeks to First Observed Tumor | 90 | 80 | 57 |
| Liver: Hepatocellular Carcinoma or Hepatocellular Adenoma[b] | 0/19(0.00) | 1/49(0.02) | 4/46(0.09) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | Infinite | Infinite |
| Lower Limit | --- | 0.021 | 0.400 |
| Upper Limit | --- | Infinite | Infinite |
| Weeks to First Observed Tumor | --- | 104 | 104 |

TABLE 6 (CONCLUDED)

| TOPOGRAPHY:MORPHOLOGY | CONTROL | LOW DOSE | HIGH DOSE |
|---|---|---|---|
| Pituitary: Chromophobe Adenoma[b] | 0/10(0.00) | 2/31(0.06) | 1/26(0.04) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | Infinite | Infinite |
| Lower Limit | --- | 0.105 | 0.022 |
| Upper Limit | --- | Infinite | Infinite |
| Weeks to First Observed Tumor | --- | 104 | 104 |
| Uterus: Endometrial Stromal Polyp[b] | 0/18(0.00) | 3/48(0.06) | 0/44(0.00) |
| P Values[c] | N.S. | N.S. | N.S. |
| Relative Risk (Control)[d] | --- | Infinite | --- |
| Lower Limit | --- | 0.236 | --- |
| Upper Limit | --- | Infinite | --- |
| Weeks to First Observed Tumor | --- | 104 | --- |

[a]Treated groups received doses of 1250 or 2500 ppm in feed.

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

[c]The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d]The 95% confidence interval on the relative risk of the treated group to the control group.

In female mice the Fisher exact test comparing high dose to control for the combined incidence of alveolar/bronchiolar carcinomas or alveolar/bronchiolar adenomas had a probability level of P = 0.043, a marginal result which was not significant under the Bonferroni criterion.

The Fisher exact test comparing low dose to control in male mice indicated a significant negative association between dosage and the combined incidence of alveolar/bronchiolar carcinomas or alveolar/bronchiolar adenomas.

To provide additional insight into the possible carcinogenicity of this compound, 95 percent confidence intervals on the relative risk have been estimated and entered in the tables based upon the observed tumor incidence rates. In many of the intervals shown in Tables 5 and 6, the value one is included; this indicates the absence of statistically significant results. It should also be noted that many of the confidence intervals have an upper limit greater than one, indicating the theoretical possibility of tumor induction in mice by carbromal that could not be established under the conditions of this test.

# V. DISCUSSION

There were no significant positive associations between the concentrations of carbromal administered and mortality in rats or mice of either sex. Adequate numbers of animals in all groups survived sufficiently long to be at risk from late-developing tumors. Slight dose-related mean body weight depression was observed for male rats and for females of both species and the mean body weight among dosed male mice was lower than that for controls, indicating that the concentrations of carbromal administered to the animals in this bioassay may have approximated the maximum tolerated concentrations.

None of the statistical tests for any site in female rats or in mice of either sex indicated a significant positive association between compound administration and tumor incidence. There was a significant positive association between the concentrations administered and the incidences of adrenal pheochromocytomas in male rats; however, the Fisher exact comparisons were not significant.

Although carbromal is a mild sedative and hypnotic drug in humans, there was no indication that the rats or mice dosed with carbromal during the chronic bioassay manifested any comparable effect.

Carbromal was not carcinogenic and showed no ability to initiate carcinogenesis in a croton oil skin painting study in S strain albino male mice (Roe and Salamon, 1955).

Under the conditions of this bioassay, dietary administration of carbromal was not carcinogenic in Fischer 344 rats or B6C3F1 mice.

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# APPENDIX A

## SUMMARY OF THE INCIDENCE OF NEOPLASMS IN RATS TREATED WITH CARBROMAL

TABLE A1
SUMMARY OF THE INCIDENCE OF NEOPLAMS IN MALE RATS TREATED WITH CARBROMAL

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS NECROPSIED | 20 | 50 | 50 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 20 | 50 | 50 |
| INTEGUMENTARY SYSTEM | | | |
| *SUBCUT TISSUE | (20) | (50) | (50) |
| FIBROMA | | 1 (2%) | |
| RESPIRATORY SYSTEM | | | |
| #LUNG | (19) | (50) | (50) |
| NEOPLASM, NOS, METASTATIC | | | 1 (2%) |
| ALVEOLAR/BRONCHIOLAR ADENOMA | | 1 (2%) | |
| ALVEOLAR/BRONCHIOLAR CARCINOMA | 1 (5%) | | |
| SARCOMA, NOS | | 1 (2%) | |
| HEMATOPOIETIC SYSTEM | | | |
| *MULTIPLE ORGANS | (20) | (50) | (50) |
| MALIG.LYMPHOMA, UNDIFFER-TYPE | | | 1 (2%) |
| MYELOMONOCYTIC LEUKEMIA | | | 1 (2%) |
| GRANULOCYTIC LEUKEMIA | 3 (15%) | 5 (10%) | 1 (2%) |
| CIRCULATORY SYSTEM | | | |
| NONE | | | |
| DIGESTIVE SYSTEM | | | |
| #LIVER | (20) | (47) | (49) |
| HEPATOCELLULAR ADENOMA | 1 (5%) | | 1 (2%) |
| #PANCREAS | (18) | (45) | (44) |
| ACINAR-CELL ADENOMA | 1 (6%) | | |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE A1 (CONTINUED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| URINARY SYSTEM | | | |
| #KIDNEY | (20) | (50) | (50) |
| CARCINOMA, NOS, METASTATIC | | | 1 (2%) |
| ADENOCARCINOMA, NOS, METASTATIC | | | 1 (2%) |
| TUBULAR-CELL ADENOMA | | 1 (2%) | 1 (2%) |
| TUBULAR-CELL ADENOCARCINOMA | | | 1 (2%) |
| CYSTADENOCARCINOMA, METASTATIC | | | 1 (2%) |
| ENDOCRINE SYSTEM | | | |
| #PITUITARY | (17) | (41) | (40) |
| CHROMOPHOBE ADENOMA | | 2 (5%) | 3 (8%) |
| #ADRENAL | (19) | (49) | (46) |
| CARCINOMA,NOS | | 1 (2%) | |
| PHEOCHROMOCYTOMA | 1 (5%) | 2 (4%) | 8 (17%) |
| #THYROID | (13) | (40) | (43) |
| FOLLICULAR-CELL CARCINOMA | | 1 (3%) | 1 (2%) |
| C-CELL ADENOMA | | | 1 (2%) |
| PAPILLARY CYSTADENOMA, NOS | | | 1 (2%) |
| #PARATHYROID | (9) | (21) | (24) |
| ADENOMA, NOS | 1 (11%) | | |
| #PANCREATIC ISLETS | (18) | (45) | (44) |
| ISLET-CELL ADENOMA | | | 2 (5%) |
| ISLET-CELL CARCINOMA | | | 1 (2%) |
| REPRODUCTIVE SYSTEM | | | |
| *PREPUTIAL GLAND | (20) | (50) | (50) |
| ADENOMA, NOS | | | 1 (2%) |
| #TESTIS | (20) | (49) | (49) |
| INTERSTITIAL-CELL TUMOR | 18 (90%) | 45 (92%) | 47 (96%) |
| NERVOUS SYSTEM | | | |
| #BRAIN | (20) | (48) | (48) |
| NEOPLASM, NOS, METASTATIC | | | 1 (2%) |

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

TABLE A1 (CONTINUED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| MENINGIOMA | | | 1 (2%) |
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |
| MUSCULOSKELETAL SYSTEM | | | |
| NONE | | | |
| BODY CAVITIES | | | |
| *PERITONEUM | (20) | (50) | (50) |
| MESOTHELIOMA, NOS | | 2 (4%) | |
| *TUNICA VAGINALIS | (20) | (50) | (50) |
| MESOTHELIOMA, NOS | 1 (5%) | 1 (2%) | |
| ALL OTHER SYSTEMS | | | |
| NONE | | | |
| ANIMAL DISPOSITION SUMMARY | | | |
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| NATURAL DEATH@ | 5 | 5 | 3 |
| MORIBUND SACRIFICE | 2 | 2 | 3 |
| SCHEDULED SACRIFICE | | | |
| ACCIDENTALLY KILLED | | | |
| TERMINAL SACRIFICE | 13 | 43 | 44 |
| ANIMAL MISSING | | | |

@ INCLUDES AUTOLYZED ANIMALS

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

TABLE A1 (CONCLUDED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| TUMOR SUMMARY | | | |
| TOTAL ANIMALS WITH PRIMARY TUMORS* | 18 | 48 | 50 |
| TOTAL PRIMARY TUMORS | 27 | 63 | 72 |
| TOTAL ANIMALS WITH BENIGN TUMORS | 18 | 46 | 49 |
| TOTAL BENIGN TUMORS | 22 | 52 | 65 |
| TOTAL ANIMALS WITH MALIGNANT TUMORS | 4 | 7 | 7 |
| TOTAL MALIGNANT TUMORS | 4 | 8 | 7 |
| TOTAL ANIMALS WITH SECONDARY TUMORS# | | | 5 |
| TOTAL SECONDARY TUMORS | | | 5 |
| TOTAL ANIMALS WITH TUMORS UNCERTAIN- BENIGN OR MALIGNANT | 1 | 3 | |
| TOTAL UNCERTAIN TUMORS | 1 | 3 | |
| 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 TREATED WITH CARBROMAL

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS NECROPSIED | 20 | 50 | 50 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 20 | 50 | 50 |
| INTEGUMENTARY SYSTEM | | | |
| *SKIN | (20) | (50) | (50) |
| BASAL-CELL CARCINOMA | | 1 (2%) | |
| *SUBCUT TISSUE | (20) | (50) | (50) |
| FIBROMA | | 1 (2%) | 1 (2%) |
| FIBROSARCOMA | | 1 (2%) | |
| RESPIRATORY SYSTEM | | | |
| #LUNG | (19) | (49) | (49) |
| UNDIFFERENTIATED CARCINOMA METAS | | 1 (2%) | |
| ALVEOLAR/BRONCHIOLAR CARCINOMA | | 1 (2%) | |
| SARCOMA, NOS, METASTATIC | | 1 (2%) | |
| HEMATOPOIETIC SYSTEM | | | |
| *MULTIPLE ORGANS | (20) | (50) | (50) |
| LEUKEMIA,NOS | 1 (5%) | 2 (4%) | 1 (2%) |
| UNDIFFERENTIATED LEUKEMIA | | 1 (2%) | |
| MYELOMONOCYTIC LEUKEMIA | | | 1 (2%) |
| ERYTHROCYTIC LEUKEMIA | | | 1 (2%) |
| GRANULOCYTIC LEUKEMIA | 2 (10%) | 1 (2%) | 1 (2%) |
| CIRCULATORY SYSTEM | | | |
| NONE | | | |
| DIGESTIVE SYSTEM | | | |
| NONE | | | |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE A2 (CONTINUED)

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| URINARY SYSTEM | | | |
| NONE | | | |
| ENDOCRINE SYSTEM | | | |
| #PITUITARY | (19) | (46) | (49) |
| CHROMOPHOBE ADENOMA | 3 (16%) | 11 (24%) | 16 (33%) |
| ACIDOPHIL ADENOMA | 1 (5%) | | |
| #ADRENAL | (20) | (49) | (50) |
| PHEOCHROMOCYTOMA | 1 (5%) | 2 (4%) | |
| SARCOMA, NOS, METASTATIC | | 1 (2%) | |
| #THYROID | (17) | (36) | (35) |
| FOLLICULAR-CELL ADENOMA | | 1 (3%) | 1 (3%) |
| C-CELL ADENOMA | 1 (6%) | 1 (3%) | 2 (6%) |
| REPRODUCTIVE SYSTEM | | | |
| *MAMMARY GLAND | (20) | (50) | (50) |
| FIBROADENOMA | 3 (15%) | 3 (6%) | 2 (4%) |
| #UTERUS | (20) | (47) | (47) |
| ADENOCARCINOMA, NOS | 1 (5%) | | 1 (2%) |
| FIBROMA | | | 1 (2%) |
| ENDOMETRIAL STROMAL POLYP | 2 (10%) | 2 (4%) | 4 (9%) |
| NERVOUS SYSTEM | | | |
| NONE | | | |
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |
| MUSCULOSKELETAL SYSTEM | | | |
| NONE | | | |
| BODY CAVITIES | | | |
| NONE | | | |

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

TABLE A2 (CONCLUDED)

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| ALL OTHER SYSTEMS | | | |
| *MULTIPLE ORGANS | (20) | (50) | (50) |
| UNDIFFERENTIATED CARCINOMA | | 1 (2%) | |
| ANIMAL DISPOSITION SUMMARY | | | |
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| NATURAL DEATH@ | 3 | 4 | 2 |
| MORIBUND SACRIFICE | | 5 | 3 |
| SCHEDULED SACRIFICE | | | |
| ACCIDENTALLY KILLED | | | |
| TERMINAL SACRIFICE | 17 | 41 | 45 |
| ANIMAL MISSING | | | |
| @ INCLUDES AUTOLYZED ANIMALS | | | |
| TUMOR SUMMARY | | | |
| TOTAL ANIMALS WITH PRIMARY TUMORS* | 10 | 23 | 26 |
| TOTAL PRIMARY TUMORS | 15 | 29 | 32 |
| TOTAL ANIMALS WITH BENIGN TUMORS | 7 | 17 | 22 |
| TOTAL BENIGN TUMORS | 11 | 21 | 27 |
| TOTAL ANIMALS WITH MALIGNANT TUMORS | 4 | 7 | 5 |
| TOTAL MALIGNANT TUMORS | 4 | 8 | 5 |
| TOTAL ANIMALS WITH SECONDARY TUMORS# | | 2 | |
| TOTAL SECONDARY TUMORS | | 3 | |
| 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 B

## SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MICE TREATED WITH CARBROMAL

TABLE B1
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE TREATED WITH CARBROMAL

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS MISSING | | 14 | 1 |
| ANIMALS NECROPSIED | 20 | 35 | 49 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 20 | 35 | 49 |
| INTEGUMENTARY SYSTEM | | | |
| *SUBCUT TISSUE | (20) | (35) | (49) |
| LIPOMA | | | 1 (2%) |
| RESPIRATORY SYSTEM | | | |
| #LUNG | (20) | (34) | (46) |
| HEPATOCELLULAR CARCINOMA, METAST | | | 1 (2%) |
| ALVEOLAR/BRONCHIOLAR ADENOMA | 6 (30%) | 4 (12%) | 8 (17%) |
| ALVEOLAR/BRONCHIOLAR CARCINOMA | 1 (5%) | | |
| HEMATOPOIETIC SYSTEM | | | |
| *MULTIPLE ORGANS | (20) | (35) | (49) |
| MALIGNANT LYMPHOMA, NOS | 1 (5%) | 2 (6%) | 1 (2%) |
| MALIG.LYMPHOMA, UNDIFFER-TYPE | | | 1 (2%) |
| MALIG.LYMPHOMA, HISTIOCYTIC TYPE | | 1 (3%) | |
| LYMPHOCYTIC LEUKEMIA | | | 1 (2%) |
| #SPLEEN | (20) | (35) | (44) |
| MALIG.LYMPHOMA, UNDIFFER-TYPE | | | 1 (2%) |
| #MESENTERIC L. NODE | (20) | (33) | (40) |
| MALIGNANT LYMPHOMA, NOS | 2 (10%) | | 1 (3%) |
| CIRCULATORY SYSTEM | | | |
| NONE | | | |
| DIGESTIVE SYSTEM | | | |
| #LIVER | (20) | (35) | (48) |
| HEPATOCELLULAR ADENOMA | 1 (5%) | 2 (6%) | 7 (15%) |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE B1 (CONTINUED)

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| HEPATOCELLULAR CARCINOMA | 3 (15%) | 6 (17%) | 6 (13%) |
| HEMANGIOMA | | 1 (3%) | |
| #PANCREAS | (20) | (35) | (47) |
| FIBROMA | 1 (5%) | | |
| URINARY SYSTEM | | | |
| NONE | | | |
| ENDOCRINE SYSTEM | | | |
| NONE | | | |
| REPRODUCTIVE SYSTEM | | | |
| NONE | | | |
| NERVOUS SYSTEM | | | |
| #BRAIN | (20) | (33) | (47) |
| ASTROCYTOMA | | 1 (3%) | |
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |
| MUSCULOSKELETAL SYSTEM | | | |
| NONE | | | |
| BODY CAVITIES | | | |
| NONE | | | |
| ALL OTHER SYSTEMS | | | |
| NONE | | | |

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

TABLE B1 (CONCLUDED)

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| ANIMAL DISPOSITION SUMMARY | | | |
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| NATURAL DEATH@ | 1 | 2 | 3 |
| MORIBUND SACRIFICE | 1 | 3 | 5 |
| SCHEDULED SACRIFICE | | | |
| ACCIDENTALLY KILLED | | | |
| TERMINAL SACRIFICE | 18 | 30 | 41 |
| ANIMAL MISSING | | 14 | 1 |
| @ INCLUDES AUTOLYZED ANIMALS | | | |
| TUMOR SUMMARY | | | |
| TOTAL ANIMALS WITH PRIMARY TUMORS* | 12 | 16 | 21 |
| TOTAL PRIMARY TUMORS | 15 | 17 | 27 |
| TOTAL ANIMALS WITH BENIGN TUMORS | 8 | 7 | 16 |
| TOTAL BENIGN TUMORS | 8 | 7 | 16 |
| TOTAL ANIMALS WITH MALIGNANT TUMORS | 6 | 10 | 10 |
| TOTAL MALIGNANT TUMORS | 7 | 10 | 11 |
| TOTAL ANIMALS WITH SECONDARY TUMORS# | | | 1 |
| TOTAL SECONDARY TUMORS | | | 1 |
| 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 TREATED WITH CARBROMAL

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS MISSING | 1 | 1 | 2 |
| ANIMALS NECROPSIED | 19 | 49 | 47 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 19 | 49 | 47 |
| INTEGUMENTARY SYSTEM | | | |
| NONE | | | |
| RESPIRATORY SYSTEM | | | |
| #LUNG | (18) | (49) | (47) |
| ALVEOLAR/BRONCHIOLAR ADENOMA | | 7 (14%) | 8 (17%) |
| ALVEOLAR/BRONCHIOLAR CARCINOMA | | 2 (4%) | 1 (2%) |
| OSTEOSARCOMA, METASTATIC | | 1 (2%) | |
| HEMATOPOIETIC SYSTEM | | | |
| *MULTIPLE ORGANS | (19) | (49) | (47) |
| MALIGNANT LYMPHOMA, NOS | 3 (16%) | 10 (20%) | 5 (11%) |
| MALIG.LYMPHOMA, UNDIFFER-TYPE | | | 1 (2%) |
| LEUKEMIA,NOS | | 1 (2%) | 1 (2%) |
| UNDIFFERENTIATED LEUKEMIA | | 1 (2%) | |
| LYMPHOCYTIC LEUKEMIA | 1 (5%) | 3 (6%) | |
| GRANULOCYTIC LEUKEMIA | | | 1 (2%) |
| #SPLEEN | (17) | (47) | (46) |
| MALIG.LYMPHOMA, HISTIOCYTIC TYPE | 1 (6%) | | 1 (2%) |
| GRANULOCYTIC LEUKEMIA | | 1 (2%) | |
| #LYMPH NODE | (16) | (45) | (37) |
| MALIGNANT LYMPHOMA, NOS | | 1 (2%) | |
| #MESENTERIC L. NODE | (16) | (45) | (37) |
| SARCOMA, NOS | | 1 (2%) | |
| MALIGNANT LYMPHOMA, NOS | | 1 (2%) | 2 (5%) |
| #LIVER | (19) | (49) | (46) |
| MALIGNANT LYMPHOMA, NOS | | | 1 (2%) |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE B2 (CONTINUED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| #SMALL INTESTINE | (18) | (48) | (44) |
| MALIGNANT LYMPHOMA, NOS | 1 (6%) | | |
| #KIDNEY | (19) | (49) | (46) |
| MALIGNANT LYMPHOMA, NOS | | | 1 (2%) |
| CIRCULATORY SYSTEM | | | |
| NONE | | | |
| DIGESTIVE SYSTEM | | | |
| #LIVER | (19) | (49) | (46) |
| HEPATOCELLULAR ADENOMA | | 1 (2%) | 2 (4%) |
| HEPATOCELLULAR CARCINOMA | | | 2 (4%) |
| URINARY SYSTEM | | | |
| NONE | | | |
| ENDOCRINE SYSTEM | | | |
| #PITUITARY | (10) | (31) | (26) |
| CHROMOPHOBE ADENOMA | | 2 (6%) | 1 (4%) |
| #PARATHYROID | (8) | (12) | (14) |
| ADENOMA, NOS | 1 (13%) | | |
| REPRODUCTIVE SYSTEM | | | |
| #UTERUS | (18) | (48) | (44) |
| LEIOMYOMA | | | 1 (2%) |
| ENDOMETRIAL STROMAL POLYP | | 3 (6%) | |
| #OVARY | (6) | (24) | (23) |
| CYSTADENOMA, NOS | | 1 (4%) | |
| NERVOUS SYSTEM | | | |
| NONE | | | |

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

TABLE B2 (CONTINUED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |
| MUSCULOSKELETAL SYSTEM | | | |
| NONE | | | |
| BODY CAVITIES | | | |
| *PELVIS | (19) | (49) | (47) |
| OSTEOSARCOMA | | 1 (2%) | |
| ALL OTHER SYSTEMS | | | |
| NONE | | | |
| ANIMAL DISPOSITION SUMMARY | | | |
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| NATURAL DEATH@ | 4 | 7 | 6 |
| MORIBUND SACRIFICE | | 6 | |
| SCHEDULED SACRIFICE | | | |
| ACCIDENTALLY KILLED | | | |
| TERMINAL SACRIFICE | 15 | 36 | 41 |
| ANIMAL MISSING | 1 | 1 | 2 |

@ INCLUDES AUTOLYZED ANIMALS

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

TABLE B2 (CONCLUDED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| TUMOR SUMMARY | | | |
| TOTAL ANIMALS WITH PRIMARY TUMORS* | 7 | 30 | 25 |
| TOTAL PRIMARY TUMORS | 7 | 36 | 28 |
| TOTAL ANIMALS WITH BENIGN TUMORS | 1 | 13 | 12 |
| TOTAL BENIGN TUMORS | 1 | 14 | 12 |
| TOTAL ANIMALS WITH MALIGNANT TUMORS | 6 | 20 | 16 |
| TOTAL MALIGNANT TUMORS | 6 | 22 | 16 |
| TOTAL ANIMALS WITH SECONDARY TUMORS# | | 1 | |
| TOTAL SECONDARY TUMORS | | 1 | |
| 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 TREATED WITH CARBROMAL

TABLE C1
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS TREATED WITH CARBROMAL

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS NECROPSIED | 20 | 50 | 50 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 20 | 50 | 50 |
| INTEGUMENTARY SYSTEM | | | |
| *SKIN | (20) | (50) | (50) |
| DERMAL INCLUSION CYST | 1 (5%) | 1 (2%) | |
| RESPIRATORY SYSTEM | | | |
| #LUNG | (19) | (50) | (50) |
| MINERALIZATION | | | 1 (2%) |
| MUCOCELE | | 1 (2%) | |
| ATELECTASIS | | 1 (2%) | 1 (2%) |
| CONGESTION, NOS | 1 (5%) | 7 (14%) | 8 (16%) |
| HEMORRHAGE | | 1 (2%) | 1 (2%) |
| PNEUMONIA, CHRONIC MURINE | 11 (58%) | 38 (76%) | 41 (82%) |
| GRANULOMA, NOS | | | 1 (2%) |
| FIBROSIS, FOCAL | | | 1 (2%) |
| FOAM-CELL | 1 (5%) | 1 (2%) | 4 (8%) |
| HYPERPLASIA, ADENOMATOUS | 2 (11%) | 1 (2%) | 1 (2%) |
| HEMATOPOIETIC SYSTEM | | | |
| #SPLEEN | (20) | (47) | (49) |
| HEMORRHAGE | | | 1 (2%) |
| #SPLENIC CAPSULE | (20) | (47) | (49) |
| FIBROSIS | | | 1 (2%) |
| HYPERPLASIA, RETICULUM CELL | | | 1 (2%) |
| CIRCULATORY SYSTEM | | | |
| #HEART/ATRIUM | (17) | (48) | (50) |
| THROMBUS, ORGANIZED | | 1 (2%) | |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE C1 (CONTINUED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| #MYOCARDIUM | (17) | (48) | (50) |
| FIBROSIS | 15 (88%) | 22 (46%) | 32 (64%) |
| FIBROSIS, FOCAL | | | 1 (2%) |
| DEGENERATION, NOS | | 2 (4%) | 1 (2%) |
| *PULMONARY ARTERY | (20) | (50) | (50) |
| EMBOLISM, NOS | | 1 (2%) | |
| DIGESTIVE SYSTEM | | | |
| #SALIVARY GLAND | (19) | (46) | (47) |
| FIBROSIS | | 1 (2%) | |
| ATROPHY, FOCAL | | 1 (2%) | |
| HYPERTROPHY, NOS | | 1 (2%) | |
| #LIVER | (20) | (47) | (49) |
| CONGESTION, NOS | | 5 (11%) | 6 (12%) |
| GRANULOMA, NOS | | | 2 (4%) |
| DEGENERATION, NOS | 1 (5%) | 3 (6%) | 1 (2%) |
| DEGENERATION, GRANULAR | | | 1 (2%) |
| METAMORPHOSIS FATTY | | 1 (2%) | 10 (20%) |
| LIPOIDOSIS | | 1 (2%) | |
| GLYCOGENIC CELL | | | 1 (2%) |
| HYPERPLASIA, FOCAL | 10 (50%) | 6 (13%) | 3 (6%) |
| #LIVER/CENTRILOBULAR | (20) | (47) | (49) |
| CONGESTION, NOS | | 1 (2%) | 3 (6%) |
| METAMORPHOSIS FATTY | 1 (5%) | 1 (2%) | 10 (20%) |
| #LIVER/HEPATOCYTES | (20) | (47) | (49) |
| HYPERPLASIA, NOS | 1 (5%) | | |
| #BILE DUCT | (20) | (47) | (49) |
| HYPERPLASIA, NOS | 8 (40%) | 7 (15%) | 8 (16%) |
| #PANCREAS | (18) | (45) | (44) |
| ATROPHY, NOS | | | 1 (2%) |
| ATROPHY, FOCAL | 2 (11%) | 5 (11%) | 2 (5%) |
| ATROPHY, DIFFUSE | 1 (6%) | | |
| #LARGE INTESTINE | (19) | (48) | (47) |
| NEMATODIASIS | | 1 (2%) | |
| PARASITISM | 8 (42%) | 16 (33%) | 7 (15%) |

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

TABLE C1 (CONTINUED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| URINARY SYSTEM | | | |
| #KIDNEY | (20) | (50) | (50) |
| MINERALIZATION | | 1 (2%) | 1 (2%) |
| HYDRONEPHROSIS | | 1 (2%) | |
| INFLAMMATION, FOCAL | | 1 (2%) | |
| INFLAMMATION, CHRONIC | 16 (80%) | 43 (86%) | 42 (84%) |
| NEPHROPATHY, TOXIC | | | 3 (6%) |
| NEPHROSIS, NOS | | 1 (2%) | |
| HYPERPLASIA, EPITHELIAL | | | 1 (2%) |
| HYPERPLASIA, PAPILLARY | | 1 (2%) | |
| LYMPHOCYTOSIS | | 1 (2%) | |
| #KIDNEY/MEDULLA | (20) | (50) | (50) |
| MINERALIZATION | | | 4 (8%) |
| #RENAL PAPILLA | (20) | (50) | (50) |
| MINERALIZATION | | 1 (2%) | |
| #KIDNEY/TUBULE | (20) | (50) | (50) |
| NECROSIS, NOS | 1 (5%) | 1 (2%) | |
| CALCIFICATION, NOS | | | 1 (2%) |
| #KIDNEY/PELVIS | (20) | (50) | (50) |
| CALCIFICATION, NOS | | 1 (2%) | |
| HYPERPLASIA, EPITHELIAL | 1 (5%) | 2 (4%) | 7 (14%) |
| #URINARY BLADDER | (17) | (32) | (33) |
| INFLAMMATION, NOS | | 1 (3%) | |
| INFLAMMATION, HEMORRHAGIC | 1 (6%) | | |
| INFLAMMATION, ACUTE | | | 1 (3%) |
| ENDOCRINE SYSTEM | | | |
| #PITUITARY | (17) | (41) | (40) |
| CYST, NOS | | | 1 (3%) |
| #ADRENAL | (19) | (49) | (46) |
| DILATATION/SINUS | | 1 (2%) | |
| HEMORRHAGIC CYST | | 1 (2%) | |
| #ADRENAL CORTEX | (19) | (49) | (46) |
| HYPERPLASIA, FOCAL | | | 1 (2%) |

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

TABLE C1 (CONTINUED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| #THYROID | (13) | (40) | (43) |
| HYPERPLASIA, C-CELL | 1 (8%) | | 2 (5%) |
| #THYROID FOLLICLE | (13) | (40) | (43) |
| CYST, NOS | 1 (8%) | | |
| HYPERTROPHY, NOS | | 1 (3%) | |
| HYPERPLASIA, CYSTIC | | 2 (5%) | 1 (2%) |
| #PANCREATIC ISLETS | (18) | (45) | (44) |
| HYPERTROPHY, NOS | | 1 (2%) | |
| REPRODUCTIVE SYSTEM | | | |
| #PROSTATE | (18) | (29) | (29) |
| INFLAMMATION, ACUTE | | | 1 (3%) |
| *SEMINAL VESICLE | (20) | (50) | (50) |
| OBSTRUCTION, NOS | | 1 (2%) | 1 (2%) |
| INFLAMMATION, CHRONIC | | | 1 (2%) |
| #TESTIS | (20) | (49) | (49) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | | 1 (2%) | |
| *EPIDIDYMIS | (20) | (50) | (50) |
| INFLAMMATION, ACUTE | | 1 (2%) | |
| NERVOUS SYSTEM | | | |
| #BRAIN | (20) | (48) | (48) |
| HEMORRHAGE | | 1 (2%) | 1 (2%) |
| ABSCESS, NOS | | 1 (2%) | 1 (2%) |
| #CEREBELLUM | (20) | (48) | (48) |
| HEMORRHAGE | 1 (5%) | | |
| MALACIA | 1 (5%) | | |
| SPECIAL SENSE ORGANS | | | |
| *EYE | (20) | (50) | (50) |
| ABSCESS, NOS | | 1 (2%) | |
| *EYE/LACRIMAL GLAND | (20) | (50) | (50) |
| ATROPHY, NOS | | 1 (2%) | |

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

TABLE C1 (CONCLUDED)

| | CONTROL(UNTR) 11-1355 | LOW DOSE 11-1353 | HIGH DOSE 11-1351 |
|---|---|---|---|
| *EYE/LACRIMAL DUCT | (20) | (50) | (50) |
| OBSTRUCTION, NOS | | 1 (2%) | |
| MUSCULOSKELETAL SYSTEM | | | |
| NONE | | | |
| BODY CAVITIES | | | |
| *MESENTERY | (20) | (50) | (50) |
| NECROSIS, FAT | | | 1 (2%) |
| ALL OTHER SYSTEMS | | | |
| ADIPOSE TISSUE | | | |
| ABSCESS, NOS | | 1 | |
| NECROSIS, NOS | | | 1 |
| SPECIAL MORPHOLOGY SUMMARY | | | |
| NO LESION REPORTED | 1 | | |
| AUTO/NECROPSY/HISTO PERF | 1 | | |

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

TABLE C2
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS TREATED WITH CARBROMAL

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS NECROPSIED | 20 | 50 | 50 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 20 | 50 | 50 |
| INTEGUMENTARY SYSTEM | | | |
| *SKIN | (20) | (50) | (50) |
| DERMAL INCLUSION CYST | 1 (5%) | | |
| *SUBCUT TISSUE | (20) | (50) | (50) |
| CYST, NOS | | 1 (2%) | |
| RESPIRATORY SYSTEM | | | |
| *NASAL TURBINATE | (20) | (50) | (50) |
| HEMORRHAGE | | | 1 (2%) |
| #TRACHEA | (15) | (40) | (44) |
| INFLAMMATION, NOS | | 2 (5%) | |
| INFLAMMATION, CHRONIC | | 1 (3%) | |
| #LUNG | (19) | (49) | (49) |
| ATELECTASIS | | 2 (4%) | 1 (2%) |
| CONGESTION, NOS | 1 (5%) | 3 (6%) | 1 (2%) |
| ABSCESS, NOS | | 1 (2%) | |
| PNEUMONIA, CHRONIC MURINE | 15 (79%) | 30 (61%) | 43 (88%) |
| GRANULOMA, FOREIGN BODY | | | 1 (2%) |
| FOAM-CELL | 1 (5%) | 7 (14%) | 14 (29%) |
| HYPERPLASIA, ADENOMATOUS | 2 (11%) | | 1 (2%) |
| HEMATOPOIETIC SYSTEM | | | |
| #SPLEEN | (18) | (45) | (50) |
| INFARCT, NOS | | | 1 (2%) |
| HEMOSIDEROSIS | | 1 (2%) | 1 (2%) |
| VASCULARIZATION | 1 (6%) | | |
| HEMATOPOIESIS | | 2 (4%) | |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE C2 (CONTINUED)

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| #MANDIBULAR L. NODE | (20) | (45) | (50) |
| MASTOCYTOSIS | | 1 (2%) | |
| #MESENTERIC L. NODE | (20) | (45) | (50) |
| ABSCESS, NOS | | | 1 (2%) |
| MASTOCYTOSIS | | 1 (2%) | |
| CIRCULATORY SYSTEM | | | |
| #MYOCARDIUM | (20) | (48) | (50) |
| MINERALIZATION | | 1 (2%) | |
| INFLAMMATION, FOCAL | | | 1 (2%) |
| INFLAMMATION, CHRONIC FOCAL | | 1 (2%) | |
| FIBROSIS | 12 (60%) | 10 (21%) | 20 (40%) |
| FIBROSIS, FOCAL | | 1 (2%) | 1 (2%) |
| #ENDOCARDIUM | (20) | (48) | (50) |
| FIBROSIS | | | 1 (2%) |
| *BLOOD VESSEL | (20) | (50) | (50) |
| MINERALIZATION | | 1 (2%) | |
| DIGESTIVE SYSTEM | | | |
| #LIVER | (20) | (48) | (50) |
| GRANULOMA, NOS | | | 4 (8%) |
| DEGENERATION, NOS | | 2 (4%) | 2 (4%) |
| NECROSIS, FOCAL | | | 1 (2%) |
| METAMORPHOSIS FATTY | | | 7 (14%) |
| CYTOPLASMIC CHANGE, NOS | | | 1 (2%) |
| BASOPHILIC CYTO CHANGE | | | 1 (2%) |
| HYPERPLASIA, FOCAL | 15 (75%) | 16 (33%) | 21 (42%) |
| HEMATOPOIESIS | | 1 (2%) | |
| #LIVER/CENTRILOBULAR | (20) | (48) | (50) |
| NECROSIS, NOS | | 1 (2%) | |
| METAMORPHOSIS FATTY | | | 11 (22%) |
| #BILE DUCT | (20) | (48) | (50) |
| HYPERPLASIA, NOS | 3 (15%) | 5 (10%) | 9 (18%) |
| #PANCREAS | (19) | (46) | (49) |
| FIBROSIS, DIFFUSE | | 1 (2%) | |

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

TABLE C2 (CONTINUED)

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| ATROPHY, FOCAL | | 5 (11%) | 3 (6%) |
| #STOMACH | (20) | (50) | (49) |
| MINERALIZATION | | 1 (2%) | |
| CYST, NOS | | | 1 (2%) |
| #LARGE INTESTINE | (19) | (46) | (48) |
| PARASITISM | 10 (53%) | 5 (11%) | 13 (27%) |
| **URINARY SYSTEM** | | | |
| #KIDNEY | (20) | (49) | (50) |
| MINERALIZATION | | 1 (2%) | |
| HYDRONEPHROSIS | | 1 (2%) | |
| INFLAMMATION, CHRONIC | 16 (80%) | 26 (53%) | 42 (84%) |
| NEPHROPATHY, TOXIC | | 1 (2%) | |
| #KIDNEY/TUBULE | (20) | (49) | (50) |
| DEGENERATION, NOS | | 2 (4%) | 1 (2%) |
| NECROSIS, NOS | 5 (25%) | 1 (2%) | 1 (2%) |
| #URINARY BLADDER | (15) | (25) | (40) |
| HEMORRHAGE | | | 1 (3%) |
| INFLAMMATION, ACUTE | 1 (7%) | | |
| **ENDOCRINE SYSTEM** | | | |
| #PITUITARY | (19) | (46) | (49) |
| CYST, NOS | 3 (16%) | 2 (4%) | 1 (2%) |
| PIGMENTATION, NOS | | 1 (2%) | |
| #ADRENAL | (20) | (49) | (50) |
| LIPOIDOSIS | | | 2 (4%) |
| CYTOPLASMIC VACUOLIZATION | | 1 (2%) | |
| #ADRENAL CORTEX | (20) | (49) | (50) |
| LIPOIDOSIS | | 1 (2%) | |
| #THYROID | (17) | (36) | (35) |
| HYPERPLASIA, C-CELL | 1 (6%) | 1 (3%) | 1 (3%) |
| HYPERPLASIA, FOLLICULAR-CELL | | 1 (3%) | |
| **REPRODUCTIVE SYSTEM** | | | |
| +MAMMARY GLAND | (20) | (50) | (50) |
| DILATATION/DUCTS | 3 (15%) | 1 (2%) | 2 (4%) |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
+ NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONTINUED)

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| *CLITORAL GLAND | (20) | (50) | (50) |
| ABSCESS, NOS | | | 1 (2%) |
| #UTERUS | (20) | (47) | (47) |
| DILATATION, NOS | | | 1 (2%) |
| CYST, NOS | | | 1 (2%) |
| INFLAMMATION, ACUTE | | 1 (2%) | |
| ABSCESS, NOS | 2 (10%) | 1 (2%) | 2 (4%) |
| FIBROSIS | | 1 (2%) | |
| NECROSIS, NOS | | 1 (2%) | 1 (2%) |
| AMYLOIDOSIS | | | 1 (2%) |
| #CERVIX UTERI | (20) | (47) | (47) |
| CYST, NOS | 1 (5%) | | 1 (2%) |
| EPIDERMAL INCLUSION CYST | | | 1 (2%) |
| HEMORRHAGE | | | 1 (2%) |
| INFLAMMATION, ACUTE | | 1 (2%) | 3 (6%) |
| ABSCESS, NOS | | 1 (2%) | |
| INFLAMMATION, ACUTE/CHRONIC | | 1 (2%) | |
| #UTERUS/ENDOMETRIUM | (20) | (47) | (47) |
| INFLAMMATION, NOS | 7 (35%) | 4 (9%) | 7 (15%) |
| INFLAMMATION, ACUTE | 4 (20%) | 1 (2%) | 6 (13%) |
| ABSCESS, NOS | 2 (10%) | | |
| HYPERPLASIA, NOS | 4 (20%) | 2 (4%) | 1 (2%) |
| HYPERPLASIA, CYSTIC | 4 (20%) | | 5 (11%) |
| #OVARY/OVIDUCT | (20) | (47) | (47) |
| INFLAMMATION, ACUTE | 1 (5%) | | |
| #OVARY | (20) | (44) | (47) |
| FOLLICULAR CYST, NOS | 1 (5%) | | 3 (6%) |
| ABSCESS, NOS | 1 (5%) | | |
| NERVOUS SYSTEM | | | |
| #BRAIN | (18) | (47) | (49) |
| INFLAMMATION ACUTE AND CHRONIC | | | 1 (2%) |
| MALACIA | | | 1 (2%) |
| #CEREBELLUM | (18) | (47) | (49) |
| HEMORRHAGE | | | 1 (2%) |
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |

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

TABLE C2 (CONCLUDED)

| | CONTROL(UNTR) 11-1356 | LOW DOSE 11-1354 | HIGH DOSE 11-1352 |
|---|---|---|---|
| MUSCULOSKELETAL SYSTEM | | | |
| *STERNUM | (20) | (50) | (50) |
| ABSCESS, NOS | | 1 (2%) | |
| BODY CAVITIES | | | |
| *SUBPLEURAL TISSUE | (20) | (50) | (50) |
| FOAM-CELL | | 1 (2%) | |
| ALL OTHER SYSTEMS | | | |
| ADIPOSE TISSUE | | | |
| NECROSIS, NOS | | 1 | |
| SPECIAL MORPHOLOGY SUMMARY | | | |
| NO LESION REPORTED | 1 | 5 | |

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

# APPENDIX D

# SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MICE TREATED WITH CARBROMAL

TABLE D1
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE TREATED WITH CARBROMAL

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS MISSING | | 14 | 1 |
| ANIMALS NECROPSIED | 20 | 35 | 49 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 20 | 35 | 49 |
| INTEGUMENTARY SYSTEM | | | |
| *SUBCUT TISSUE | (20) | (35) | (49) |
| HEMATOMA, NOS | | 1 (3%) | |
| RESPIRATORY SYSTEM | | | |
| #LUNG/BRONCHUS | (20) | (34) | (46) |
| HYPERPLASIA, EPITHELIAL | 1 (5%) | | |
| #LUNG | (20) | (34) | (46) |
| CONGESTION, NOS | | 1 (3%) | |
| BRONCHOPNEUMONIA, ACUTE | | | 1 (2%) |
| PNEUMONIA, CHRONIC MURINE | | 1 (3%) | 1 (2%) |
| HYPERPLASIA, ADENOMATOUS | | 1 (3%) | 2 (4%) |
| HEMATOPOIETIC SYSTEM | | | |
| #BONE MARROW | (17) | (33) | (31) |
| HYPERPLASIA, HEMATOPOIETIC | | | 6 (19%) |
| HYPERPLASIA, GRANULOCYTIC | | 1 (3%) | |
| #SPLEEN | (20) | (35) | (44) |
| DEGENERATION, HYALINE | 1 (5%) | | |
| HEMOSIDEROSIS | | | 1 (2%) |
| HYPERPLASIA, RETICULUM CELL | | | 1 (2%) |
| HYPERPLASIA, LYMPHOID | | 1 (3%) | |
| HEMATOPOIESIS | | | 2 (5%) |
| #SPLENIC FOLLICLES | (20) | (35) | (44) |
| INFLAMMATION, NOS | | | 1 (2%) |
| #MESENTERIC L. NODE | (20) | (33) | (40) |
| EDEMA, NOS | | 1 (3%) | |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE D1 (CONTINUED)

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| INFLAMMATION, NOS | | | 1 (3%) |
| HYPERPLASIA, NOS | | | 1 (3%) |
| HEMATOPOIESIS | | 1 (3%) | |
| **CIRCULATORY SYSTEM** | | | |
| #MYOCARDIUM | (20) | (34) | (45) |
| INFLAMMATION, NOS | | | 1 (2%) |
| *RENAL ARTERY | (20) | (35) | (49) |
| ARTERIOSCLEROSIS, NOS | | | 1 (2%) |
| **DIGESTIVE SYSTEM** | | | |
| #SALIVARY GLAND | (20) | (33) | (44) |
| DILATATION/DUCTS | | | 1 (2%) |
| PERIVASCULAR CUFFING | | | 1 (2%) |
| #LIVER | (20) | (35) | (48) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | | 1 (3%) | |
| DEGENERATION, HYALINE | | 1 (3%) | |
| NECROSIS, FOCAL | | 1 (3%) | |
| METAMORPHOSIS FATTY | 2 (10%) | | 1 (2%) |
| HEMOSIDEROSIS | | | 1 (2%) |
| NUCLEAR ENLARGEMENT | | 1 (3%) | |
| HEPATOCYTOMEGALY | | | 5 (10%) |
| HYPERPLASIA, NODULAR | | 1 (3%) | |
| ANGIECTASIS | | 1 (3%) | 1 (2%) |
| HEMATOPOIESIS | | | 1 (2%) |
| #LIVER/PERIPORTAL | (20) | (35) | (48) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | | 1 (3%) | |
| #PEYERS PATCH | (20) | (34) | (48) |
| HYPERPLASIA, NOS | | 3 (9%) | 2 (4%) |
| HYPERPLASIA, LYMPHOID | 1 (5%) | | |
| #LARGE INTESTINE | (20) | (35) | (45) |
| INFLAMMATION, NOS | | | 1 (2%) |
| NEMATODIASIS | 8 (40%) | 12 (34%) | 12 (27%) |
| #COLON | (20) | (35) | (45) |
| NEMATODIASIS | | 1 (3%) | |

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

TABLE D1 (CONTINUED)

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| URINARY SYSTEM | | | |
| #KIDNEY | (20) | (35) | (48) |
| INFLAMMATION, CHRONIC | 1 (5%) | 1 (3%) | 1 (2%) |
| PERIVASCULAR CUFFING | | | 2 (4%) |
| ENDOCRINE SYSTEM | | | |
| #ADRENAL | (19) | (32) | (38) |
| HYPERPLASIA, ADENOMATOUS | | | 1 (3%) |
| #ADRENAL CORTEX | (19) | (32) | (38) |
| HYPERPLASIA, NOS | | 2 (6%) | |
| HYPERPLASIA, FOCAL | | | 1 (3%) |
| #ADRENAL MEDULLA | (19) | (32) | (38) |
| DEGENERATION, HYALINE | | 1 (3%) | |
| #THYROID | (20) | (29) | (39) |
| HYPERPLASIA, C-CELL | | 1 (3%) | |
| #THYROID FOLLICLE | (20) | (29) | (39) |
| HYPERTROPHY, NOS | 1 (5%) | | |
| #PANCREATIC ISLETS | (20) | (35) | (47) |
| HYPERPLASIA, NOS | 1 (5%) | | |
| REPRODUCTIVE SYSTEM | | | |
| *MAMMARY GLAND | (20) | (35) | (49) |
| CYTOPLASMIC VACUOLIZATION | | | 1 (2%) |
| #PROSTATE | (20) | (35) | (46) |
| HYPERPLASIA, NOS | | | 1 (2%) |
| *SEMINAL VESICLE | (20) | (35) | (49) |
| FIBROSIS | | | 1 (2%) |
| #TESTIS | (20) | (34) | (47) |
| INFARCT, NOS | 1 (5%) | | |
| HYPERPLASIA, INTERSTITIAL CELL | | | 1 (2%) |

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

TABLE D1 (CONTINUED)

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| NERVOUS SYSTEM | | | |
| *NERVOUS SYSTEM | (20) | (35) | (49) |
| CALCIFICATION, FOCAL | | 1 (3%) | |
| #BRAIN | (20) | (33) | (47) |
| MINERALIZATION | 1 (5%) | | 7 (15%) |
| CALCIFICATION, FOCAL | 6 (30%) | 4 (12%) | 4 (9%) |
| CYTOPLASMIC VACUOLIZATION | | 1 (3%) | |
| #CEREBRAL WHITE MATTE | (20) | (33) | (47) |
| CYTOPLASMIC VACUOLIZATION | | 1 (3%) | 12 (26%) |
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |
| MUSCULOSKELETAL SYSTEM | | | |
| NONE | | | |
| BODY CAVITIES | | | |
| *ABDOMINAL CAVITY | (20) | (35) | (49) |
| HEMATOMA, NOS | | 1 (3%) | |
| *SUBPLEURAL TISSUE | (20) | (35) | (49) |
| FOAM-CELL | | | 1 (2%) |
| *MESENTERY | (20) | (35) | (49) |
| INFLAMMATION, GRANULOMATOUS | | 1 (3%) | |
| PERIARTERITIS | | 1 (3%) | |
| ALL OTHER SYSTEMS | | | |
| NONE | | | |
| SPECIAL MORPHOLOGY SUMMARY | | | |
| NO LESION REPORTED | 4 | 3 | 6 |

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

TABLE D1 (CONCLUDED)

| | CONTROL(UNTR) 22-2355 | LOW DOSE 22-2353 | HIGH DOSE 22-2351 |
|---|---|---|---|
| ANIMAL MISSING/NO NECROPSY | | 14 | 1 |

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

TABLE D2
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE TREATED WITH CARBROMAL

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| ANIMALS INITIALLY IN STUDY | 20 | 50 | 50 |
| ANIMALS MISSING | 1 | 1 | 2 |
| ANIMALS NECROPSIED | 19 | 49 | 47 |
| ANIMALS EXAMINED HISTOPATHOLOGICALLY** | 19 | 49 | 47 |
| INTEGUMENTARY SYSTEM | | | |
| NONE | | | |
| RESPIRATORY SYSTEM | | | |
| #LUNG/BRONCHUS | (18) | (49) | (47) |
| FOREIGN BODY, NOS | | | 1 (2%) |
| PERIVASCULAR CUFFING | 1 (6%) | | |
| #LUNG | (18) | (49) | (47) |
| CONGESTION, CHRONIC PASSIVE | | 1 (2%) | |
| INFLAMMATION, FOCAL | | 1 (2%) | |
| PNEUMONIA, GIANT-CELL | | | 1 (2%) |
| PNEUMONIA, CHRONIC MURINE | 1 (6%) | 1 (2%) | 2 (4%) |
| INFLAMMATION, CHRONIC | | 1 (2%) | |
| PERIARTERITIS | | | 1 (2%) |
| ALVEOLAR MACROPHAGES | 1 (6%) | | |
| HYPERPLASIA, ADENOMATOUS | | 2 (4%) | |
| HEMATOPOIETIC SYSTEM | | | |
| #BONE MARROW | (16) | (48) | (41) |
| HYPERPLASIA, HEMATOPOIETIC | | 3 (6%) | 2 (5%) |
| #SPLEEN | (17) | (47) | (46) |
| HYPERPLASIA, NOS | | | 1 (2%) |
| HYPERPLASIA, RETICULUM CELL | | | 2 (4%) |
| HYPERPLASIA, LYMPHOID | 2 (12%) | 1 (2%) | |
| #LYMPH NODE | (16) | (45) | (37) |
| HYPERPLASIA, LYMPHOID | 1 (6%) | | |
| #MANDIBULAR L. NODE | (16) | (45) | (37) |
| HYPERPLASIA, NOS | 1 (6%) | | |

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE D2 (CONTINUED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| #BRONCHIAL LYMPH NODE | (16) | (45) | (37) |
| INFLAMMATION, NOS | 1 (6%) | | |
| #MESENTERIC L. NODE | (16) | (45) | (37) |
| INFLAMMATION, NOS | 1 (6%) | | |
| INFLAMMATION, GRANULOMATOUS | | 1 (2%) | |
| HYPERPLASIA, NOS | | | 1 (3%) |
| CIRCULATORY SYSTEM | | | |
| *PULMONARY ARTERY | (19) | (49) | (47) |
| HYPERPLASIA, NOS | 1 (5%) | | |
| DIGESTIVE SYSTEM | | | |
| #SALIVARY GLAND | (15) | (44) | (42) |
| INFLAMMATION, CHRONIC | | 1 (2%) | |
| #LIVER | (19) | (49) | (46) |
| HEMORRHAGE | | | 1 (2%) |
| FIBROSIS, FOCAL | | | 1 (2%) |
| NECROSIS, FOCAL | 2 (11%) | 1 (2%) | 1 (2%) |
| METAMORPHOSIS FATTY | | 2 (4%) | 3 (7%) |
| NUCLEAR ALTERATION | | | 1 (2%) |
| NUCLEAR ENLARGEMENT | | 1 (2%) | |
| HEMATOPOIESIS | | | 1 (2%) |
| #LIVER/PERIPORTAL | (19) | (49) | (46) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | | 1 (2%) | |
| #LIVER/HEPATOCYTES | (19) | (49) | (46) |
| NECROSIS, NOS | | 1 (2%) | |
| #BILE DUCT | (19) | (49) | (46) |
| CYST, NOS | | | 1 (2%) |
| CALCIFICATION, NOS | | | 1 (2%) |
| #PANCREAS | (16) | (47) | (46) |
| FIBROSIS | | 1 (2%) | |
| #LARGE INTESTINE | (16) | (44) | (43) |
| NEMATODIASIS | | 2 (5%) | 1 (2%) |

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

TABLE D2 (CONTINUED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| URINARY SYSTEM | | | |
| #KIDNEY | (19) | (49) | (46) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | 1 (5%) | 1 (2%) | 1 (2%) |
| METAPLASIA, OSSEOUS | | | 1 (2%) |
| #KIDNEY/CORTEX | (19) | (49) | (46) |
| CYST, NOS | 1 (5%) | | |
| #URINARY BLADDER | (16) | (45) | (43) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | 1 (6%) | | |
| ENDOCRINE SYSTEM | | | |
| #PITUITARY | (10) | (31) | (26) |
| HYPERPLASIA, NOS | | 1 (3%) | |
| HYPERPLASIA, DIFFUSE | 1 (10%) | | |
| #ADRENAL | (16) | (45) | (42) |
| LIPOIDOSIS | | | 1 (2%) |
| #ADRENAL MEDULLA | (16) | (45) | (42) |
| DEGENERATION, HYALINE | | 2 (4%) | 2 (5%) |
| #THYROID | (15) | (38) | (37) |
| COLLOID CYST | | 1 (3%) | 1 (3%) |
| INFLAMMATION, ACUTE | 1 (7%) | | |
| HYPERPLASIA, EPITHELIAL | | | 1 (3%) |
| HYPERPLASIA, FOCAL | | 1 (3%) | 1 (3%) |
| #THYROID FOLLICLE | (15) | (38) | (37) |
| HYPERTROPHY, NOS | | 1 (3%) | |
| #PANCREATIC ISLETS | (16) | (47) | (46) |
| HYPERPLASIA, NOS | 1 (6%) | 4 (9%) | 1 (2%) |
| REPRODUCTIVE SYSTEM | | | |
| *MAMMARY GLAND | (19) | (49) | (47) |
| LACTATION | | 1 (2%) | |
| *VAGINA | (19) | (49) | (47) |
| INFLAMMATION, SUPPURATIVE | | | 1 (2%) |

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

TABLE D2 (CONTINUED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| #UTERUS | (18) | (48) | (44) |
| CYST, NOS | 1 (6%) | | |
| HEMORRHAGE | | 1 (2%) | |
| PYOMETRA | 8 (44%) | 14 (29%) | 25 (57%) |
| #UTERUS/ENDOMETRIUM | (18) | (48) | (44) |
| INFLAMMATION, CHRONIC | | | 1 (2%) |
| HYPERPLASIA, CYSTIC | | | 3 (7%) |
| #OVARY/OVIDUCT | (18) | (48) | (44) |
| CYST, NOS | | 1 (2%) | |
| #OVARY | (6) | (24) | (23) |
| CYST, NOS | 1 (17%) | 5 (21%) | 4 (17%) |
| FOLLICULAR CYST, NOS | | 1 (4%) | |
| ABSCESS, NOS | | 1 (4%) | |
| NERVOUS SYSTEM | | | |
| #BRAIN | (19) | (48) | (47) |
| MINERALIZATION | 1 (5%) | | |
| HEMORRHAGIC CYST | | 1 (2%) | |
| CALCIFICATION, NOS | | 1 (2%) | |
| CALCIFICATION, FOCAL | 3 (16%) | 3 (6%) | 3 (6%) |
| CYTOPLASMIC VACUOLIZATION | 2 (11%) | | 1 (2%) |
| #CEREBRAL WHITE MATTE | (19) | (48) | (47) |
| CYTOPLASMIC VACUOLIZATION | 1 (5%) | 3 (6%) | 1 (2%) |
| SPECIAL SENSE ORGANS | | | |
| NONE | | | |
| MUSCULOSKELETAL SYSTEM | | | |
| *SKELETAL MUSCLE | (19) | (49) | (47) |
| PARASITISM | 1 (5%) | | |
| BODY CAVITIES | | | |
| *ABDOMINAL CAVITY | (19) | (49) | (47) |
| STEATITIS | | 1 (2%) | |

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

TABLE D2 (CONCLUDED)

| | CONTROL(UNTR) 22-2356 | LOW DOSE 22-2354 | HIGH DOSE 22-2352 |
|---|---|---|---|
| NECROSIS, FAT | | 1 (2%) | |
| *MESENTERY | (19) | (49) | (47) |
| THROMBOSIS, NOS | 1 (5%) | | |
| INFARCT, NOS | 1 (5%) | | |
| ALL OTHER SYSTEMS | | | |
| *MULTIPLE ORGANS | (19) | (49) | (47) |
| LYMPHOCYTIC INFLAMMATORY INFILTR | 1 (5%) | | 1 (2%) |
| AMYLOIDOSIS | | | 1 (2%) |
| OMENTUM | | | |
| LYMPHOCYTIC INFLAMMATORY INFILTR | | | 1 |
| BROAD LIGAMENT | | | |
| LYMPHOCYTIC INFLAMMATORY INFILTR | | | 2 |
| SPECIAL MORPHOLOGY SUMMARY | | | |
| NO LESION REPORTED | | 6 | 2 |
| ANIMAL MISSING/NO NECROPSY | 1 | 1 | 2 |

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

Review of the Bioassay of Carbromal* for Carcinogenicity
by the Data Evaluation/Risk Assessment Subgroup of the
Clearinghouse on Environmental Carcinogens

August 31, 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 Carbromal for carcinogenicity.

The primary reviewer said that Carbromal was not carcinogenic in rats or mice, under the conditions of test. He indicated that the experimental design was straightforward and that there was no unusual highlights to report. An increased incidence of pheochromocytomas in treated male rats was observed but was not considered significant. Based on the results of the study, the primary reviewer opined that Carbromal would not appear to pose a carcinogenic risk to humans.

The secondary reviewer agreed with the conclusion in the report that Carbromal was not carcinogenic, under the conditions of test. He noted that the mice were housed in the same room in which other compounds were under test and that too few matched control animals were employed. He concluded that Carbromal probably does not pose a hazard to man in its use as a pharmaceutical. He recommended acceptance of the report as written.

A motion was approved unanimously that the report on the bioassay of Carbromal be accepted as written.

Members present were:

Arnold L. Brown (Chairman), University of Wisconsin Medical School
Joseph Highland, Environmental Defense Fund

(Verald K. Rowe, Dow Chemical USA, submitted a written review)
Michael 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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