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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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FORWARD: This report presents the results of the bioassay of malathion 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 of 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 malathion was conducted by Gulf South Research Institute, New Iberia, Louisiana, initially under direct contract to the NCI and currently under a subcontract to Tracor Jitco, Inc., prime contractor for the NCI Carcinogenesis Bioassay Program.

The experimental design was determined by Drs. J. H. Weisburger[1,2] and R. R. Bates[1,3]; the doses were selected by Drs. T. E. Shellenberger[4,5], J. H. Weisburger, and R. R. Bates. Administration of the test chemical and observation of the laboratory animals were supervised by Drs. T. E. Shellenberger and H. P. Burchfield[4], with the technical assistance of Ms. D. H. Monceaux[4] and Mr. D. Broussard[4]. Histopathology was performed by Drs. E. Bernal[4] and B. Buratto[4] at Gulf South Research

Institute, and the diagnoses included in this report represent the interpretation of these pathologists.

Animal pathology tables and survival tables were compiled at EG&G Mason Research Institute[6]. Statistical analyses were performed by Dr. J. R. Joiner[7], using methods selected for the bioassay program by Dr. J. J. Gart[8]. Chemicals used in this bioassay were analyzed under the direction of Dr. H. P. Burchfield, and the results of the analyses were reviewed by Dr. S. S. Olin[7].

This report was prepared at Tracor Jitco[7] under the direction of NCI. Those responsible for the report at Tracor Jitco were Dr. Marshall Steinberg, Director of the Bioassay Program; Dr. L. A. Campbell, Deputy Director for Science; Dr. J. F. Robens, toxicologist; Dr. R. L. Schueler, pathologist; Dr. G. L. Miller, Ms. Y. E. Presley, and Mr. W. D. Reichardt, technical writers; and Dr. E. W. Gunberg, technical editor, assisted by Ms. P. J. Graboske.

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SUMMARY

A bioassay of technical-grade malathion for possible carcinogenicity was conducted by administering the test chemical in feed to Osborne-Mendel rats and B6C3F1 mice.

Groups of 50 rats of each sex were administered malathion at one of two doses for 80 weeks, then observed for 33 weeks. Time-weighted average doses were 4,700 or 8,150 ppm. Matched controls consisted of groups of 15 untreated rats of each sex; pooled controls consisted of the matched controls combined with 40 untreated male and 40 untreated female rats from similar bioassays of four other test chemicals. All surviving rats were killed at 108-113 weeks.

Groups of 50 mice of each sex were administered malathion at one of two doses, either 8,000 or 16,000 ppm, for 80 weeks, then observed for 14 or 15 weeks. Matched controls consisted of groups of 10 untreated mice of each sex; pooled controls consisted of the matched controls combined with 40 untreated male and 40 untreated female mice from similar bioassays of four other test chemicals. All surviving mice were killed at 94 or 95 weeks.

Mortality in either rats or mice was not significantly related to the administration of malathion. Sufficient numbers of animals were at risk in the dosed and control groups of rats and mice of each sex for development of late-appearing tumors.

In female rats, three follicular-cell carcinomas and one follicular-cell adenoma of the thyroid occurred in the high-dose group, and three follicular-cell hyperplasias occurred in the low-dose group. The incidence of these tumors showed a statistically significant ($P = 0.026$) dose-related trend; however, the results of the Fisher exact test for direct comparison between the dosed and control groups were not significant. More dosed males than dosed females had either tumors or hyperplasia of the follicular

cells of the thyroid; however, because of the higher incidence of tumors among the male controls, none of the results of the statistical tests were significant. These thyroid tumors were not considered to be associated with the administration of malathion.

In male mice, hepatocellular carcinoma occurred at the following incidences: matched controls 2/10, pooled controls 5/49, low-dose 7/48, high-dose 11/49. In addition, neoplastic nodules occurred in 3/49 pooled-control and 6/49 high-dose animals. When the combined incidence of these neoplasms in the dosed animals was compared with that of the pooled controls, the dose-related trend was P = 0.019 and the direct comparison of the high-dose group with the control group was P = 0.031. Thus, none of the direct comparisons of dosed groups with controls were significant using the Bonferroni criteria. In addition, the historical controls from this laboratory had several control groups with incidences of 35-40% hepatocellular carcinoma, rates which are comparable with the incidence of this tumor in the dosed male mice of the present study. Thus, these liver tumors are not considered to be associated with the administration of malathion. The incidences of liver tumors in dosed females were not statistically significant when compared with that in control animals.

It is concluded that under the conditions of this bioassay, there was no clear evidence of the association of the tumor incidence with the administration of malathion to Osborne-Mendel rats or B6C3F1 mice.

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I. INTRODUCTION

Malathion (CAS 121-75-5; NCI C00215) is the generic name for S-(1,2-dicarbethoxyethyl)-0,0-dimethyldithiophosphate, which is an organophosphorus insecticide and acaricide first synthesized in the United States in 1952. Malathion primarily affects the nervous system by inhibition of cholinesterase activity and subsequent accumulation of acetylcholine (Koelle, 1975). However, it has a low mammalian toxicity (Eto, 1974). Malathion is approved for a wide variety of uses as an insecticide and acaricide on field crops, fruits, nut trees, vegetables, livestock, agricultural premises, and land. Tolerances for residues of malathion have been established on many of these products (EPA, 1972).

Malathion was selected for the Carcinogenesis Testing Program because of its wide use in agriculture and the absence of adequate chronic studies of the chemical.

II. MATERIALS AND METHODS

A. Chemical

The technical-grade malathion used for the chronic study was obtained in a single batch (Lot No. SPS-10127) from American Cyanamid Company, Agricultural Division, Princeton, New Jersey. The identity of the chemical was confirmed at Gulf South Research Institute by infrared, nuclear magnetic resonance, and mass spectral analyses. Gas-liquid chromatographic analysis (electron capture detector, DC-200 column) showed a single peak, consistent with the manufacturer's specification of 95% minimum purity. No attempt was made to identify or quantitate impurities.

The chemical was stored in the original container at approximately 0°C until used.

B. Dietary Preparation

All test diets were formulated using Wayne® Lab Blox animal meal (Allied Mills, Inc., Chicago, Ill.) to which was added the required amount of malathion for each dietary concentration. The test chemical was first dissolved in a small amount of acetone (Mallinckrodt, Inc., St. Louis, Mo.), which was then added to the feed. Corn oil (Louana®, Opelousas Refinery, Opelousas, La.) was also added to the feed, primarily as a dust suppressant, and the

diets were mixed mechanically to assure homogeneity of the mixtures and evaporation of the acetone. Final diets, including those for the control groups of animals, contained corn oil equal to 2% of the final weight of feed. The diets were stored at approximately 17°C until used, but no longer than 1 week.

The stability of malathion in feed was tested by determining the concentration of the chemical in formulated diets at intervals over a 7-day period. Diets containing 1,000, 8,000, or 10,000 ppm malathion showed no change on standing at ambient temperature for this period.

As a quality control test on the accuracy of preparation of the diets, the concentration of malathion was determined in different batches of formulated diets during the chronic study. The results are summarized in Appendix G. At each dietary concentration, the mean of the analytical concentrations for the checked samples was within 2.5% of the theoretical concentration, and the coefficient of variation was never more than 5.0%. Thus, the evidence indicates that the formulated diets were prepared accurately.

C. Animals

Rats and mice of each sex, obtained through contracts of the Division of Cancer Treatment, National Cancer Institute, were

used in these bioassays. The rats were of the Osborne-Mendel strain obtained from Battelle Memorial Institute, Columbus, Ohio, and the mice were B6C3F1 hybrids obtained from Charles River Breeding Laboratories, Inc., Wilmington, Massachusetts. On arrival at the laboratory, all animals were quarantined (rats for 4 days, mice for 15 days) and were then assigned to control or dosed groups.

D. Animal Maintenance

All animals were housed in temperature- and humidity-controlled rooms. The temperature range was 22-24°C, and the relative humidity was maintained at 40-70%. The air in each room was changed 10-12 times per hour. Fluorescent light provided illumination 10 hours per day. Food and water were available *ad libitum*.

The rats were housed individually in hanging galvanized steel mesh cages, and the mice were housed in plastic cages covered with filter bonnets, five animals per cage for females, and two or three animals per cage for males. Initially, rats were transferred once per week to clean cages; later in the study, cages were changed every 2 weeks. Mice were transferred once per week to clean cages covered with filter bonnets; bedding used for the mice was Absorb-Dri® (Lab Products, Inc., Garfield N. J.).

For rats, absorbent sheets under the cages were changed three times per week. Feeder jars and water bottles were changed and sterilized three times per week.

Cages for control and dosed mice were placed on separate racks in the same room. Animal racks for both species were rotated laterally once per week; at the same time, each cage was changed to a different position in the row within the same column. Rats receiving malathion, along with their matched controls, were housed in a room by themselves. Mice receiving malathion were maintained in a room housing mice fed tetrachlorvinphos (CAS 961-11-5) and dieldrin (CAS 60-57-1), together with their respective matched controls.

E. Subchronic Studies

Subchronic feeding studies were conducted to estimate the maximum tolerated doses of malathion, on the basis of which low and high concentrations (hereinafter referred to as "low doses" and "high doses") were determined for administration in the chronic studies. In these subchronic studies, malathion was added to the animal feed in twofold increasing concentrations, ranging from 1,000 to 8,000 ppm for rats and 250 to 8,000 ppm for mice. Treated and control groups each consisted of five male and five female animals. The chemical was provided in feed to dosed

groups for 6 weeks, followed by observation for 2 weeks. Because there were no deaths in any dosed group of rats or mice during the study, a second study was performed with doses of 8,000 to 32,000 ppm for rats and 4,000 to 32,000 ppm for mice.

In the second study, all rats and four male mice receiving 32,000 ppm died by week 3. No animal receiving 16,000 ppm died, but mean weight gains decreased early in the study. The low and high doses for the chronic studies using both rats and mice were set at 8,000 and 16,000 ppm.

F. Designs of Chronic Studies

The designs of the chronic studies are shown in tables 1 and 2. The matched-control groups for restarted high-dose rats included five animals of each sex; initially, 16,000 ppm was fed to rats of each sex as the high dose, and 10 matched controls of each sex were used. Because this dose of malathion was too toxic, the high-dose study was terminated and new high-dose groups were started, as shown in table 1.

Since the numbers of animals in the matched-control groups were small, pooled-control groups also were used for statistical comparisons. The pooled-control groups consisted of the matched controls from the bioassay of malathion combined with matched controls from the bioassays of tetrachlorvinphos, toxaphene (CAS

Table 1. Design of Malathion Chronic Feeding Studies in Rats

Sex and Test Group	Initial No. of Animals[a]	Malathion in Diet[b] (ppm)	Time on Study: Dosed (weeks)	Time on Study: Observed[c] (weeks)	Time-Weighted Average Dose[d] (ppm)
Male					
Low-Dose Matched-Control	10	0		113	
High-Dose Matched-Control	5	0		108	
Low-Dose	50	8,000	14		4,700
		4,000	66		
		0		33	
High-Dose[e]	50	12,000	3		
		8,000	77		8,150
		0		29	
Female					
Low-Dose Matched-Control	10	0		113	
High-Dose Matched-Control	5	0		109	
Low-Dose	50	8,000	14		4,700
		4,000	66		
		0		33	
High-Dose	50[e]	12,000	3		
		8,000	77		8,150
		0		29	

[a]All animals were 35 days of age when placed on study.

[b]Doses were lowered after 3 and 14 weeks on study, based on the pattern of mortality, changes in body weight, and the general condition of the animals in this and other carcinogen bioassays; it was believed that excessive mortality might otherwise occur before termination of the study.

Table 1. Design of Malathion Chronic Feeding Studies in Rats

(continued)

[c]When diets containing malathion were discontinued, low-dose rats and their matched controls were fed a control diet without corn oil for 2 weeks, then the control diet (2% corn oil added) for an additional 31 weeks. High-dose rats received the control diet until termination of the study.

[d]Time-weighted average dose = $\frac{\Sigma\,(\text{dose in ppm x no. of weeks at that dose})}{\Sigma\,(\text{no. of weeks receiving each dose})}$

[e]Original high-dose groups were terminated due to the toxic effects of malathion, and new high-dose groups were started at the doses indicated.

Table 2. Design of Malathion Chronic Feeding Studies in Mice

Sex and Test Group	Initial No. of Animals[a]	Malathion in Diet (ppm)	Time on Study	
			Dosed (weeks)	Observed[b] (weeks)
Male				
Matched-Control	10	0		95
Low-Dose	50	8,000	80	
		0		14-15
High-Dose	50	16,000	80	
		0		15
Female				
Matched-Control	10	0		95
Low-Dose	50	8,000	80	
		0		14
High-Dose	50	16,000	80	
		0		15

[a]All animals were 35 days of age when placed on study.

[b]When diets containing malathion were discontinued, all mice received the control diet (2% corn oil added) until termination of the study.

8001-35-2), endrin (CAS 72-20-8), and lindane (CAS 58-89-9), to give groups of 55 male and 55 female rats, and 50 male and 50 female mice. The bioassays of chemicals other than malathion were also conducted at Gulf South Research Institute and overlapped the bioassay of malathion by at least 1 year. The matched-controls were of the same strain and from the same supplier and were examined by the same pathologists. Because additional matched controls were started simultaneously with restarted dosed groups for some of these bioassays, the number of animals in the pooled-control groups varied.

G. Clinical and Pathologic Examinations

All animals were observed twice per day for signs of toxicity, weighed at regular intervals, and palpated for masses at each weighing. Animals that were moribund at the time of daily examination were killed 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, lungs and bronchi, trachea, bone and bone marrow, spleen, lymph nodes, heart, salivary gland, liver, gallbladder (mice), pancreas, stomach, small intestine, large intestine, kidney, urinary bladder,

pituitary, adrenal, thyroid, parathyroid, mammary gland, prostate or uterus, testis or ovary, and brain. 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 were 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. 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.

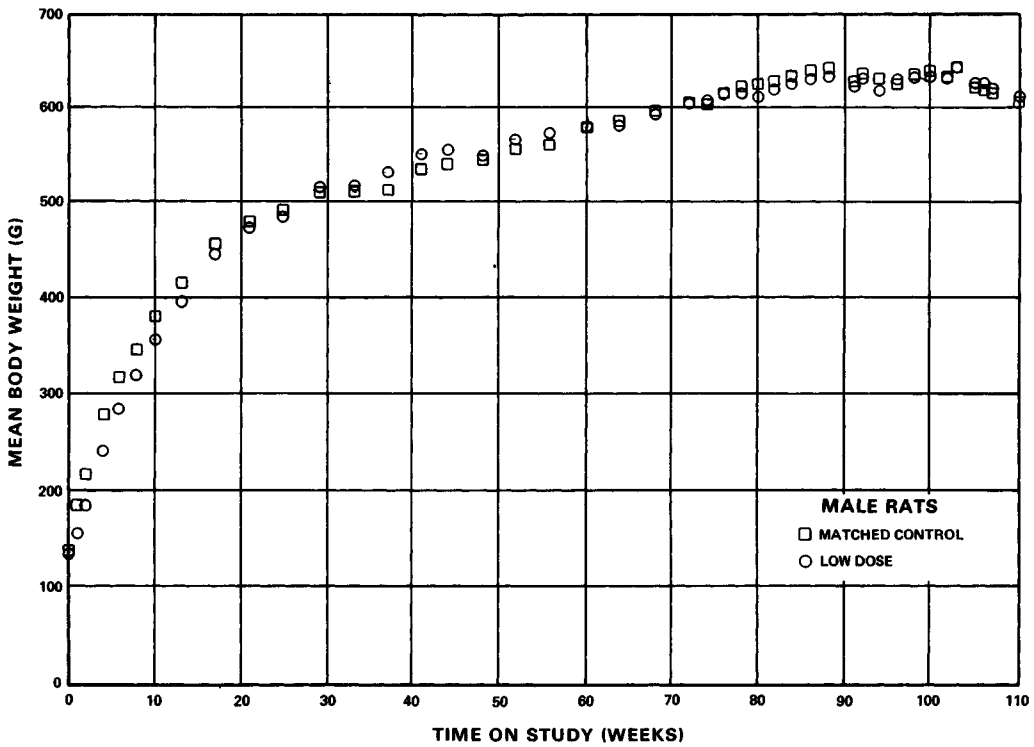
III. RESULTS - RATS

A. Body Weights and Clinical Signs (Rats)

During the period of rapid growth, the mean body weights of the low- and high-dose male rats were slightly lower than those of the controls (figure 1); thereafter, no differences between weights of dosed and control groups were discernible. In females, the mean body weights were generally lower in the dosed groups than in the controls throughout the study (figure 2). Fluctuations in the growth curve may be due to mortality; as the size of a group diminishes, the mean body weight may be subject to wide variation.

After 2 weeks on study, generalized body tremors were observed in one low-dose male and one high-dose male. During the remainder of the first year, the dosed animals were generally comparable to the controls in appearance and behavior. A few animals showed evidence of an eye infection that was diagnosed by the pathologists at the laboratory as viral conjunctivitis.

During the second year of study, clinical signs including rough hair coats, pale mucous membranes, dermatitis, ataxia, alopecia, hematuria, and vaginal bleeding were observed with increasing frequency in dosed animals.



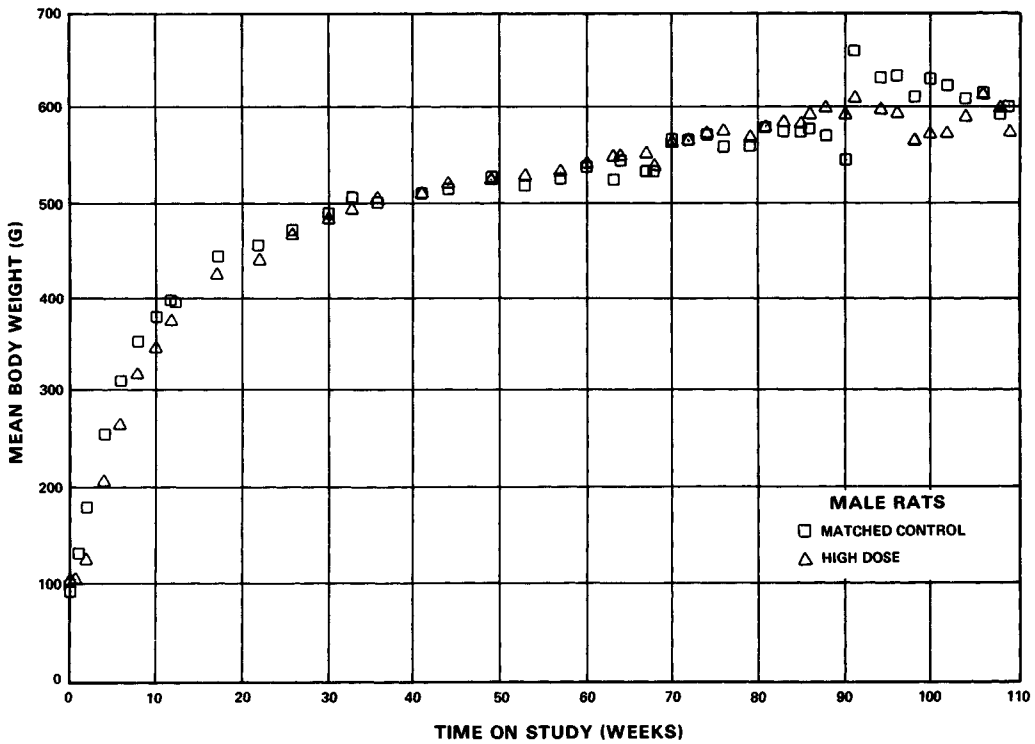


Figure 1. Growth Curves for Male Rats Fed Malathion in the Die

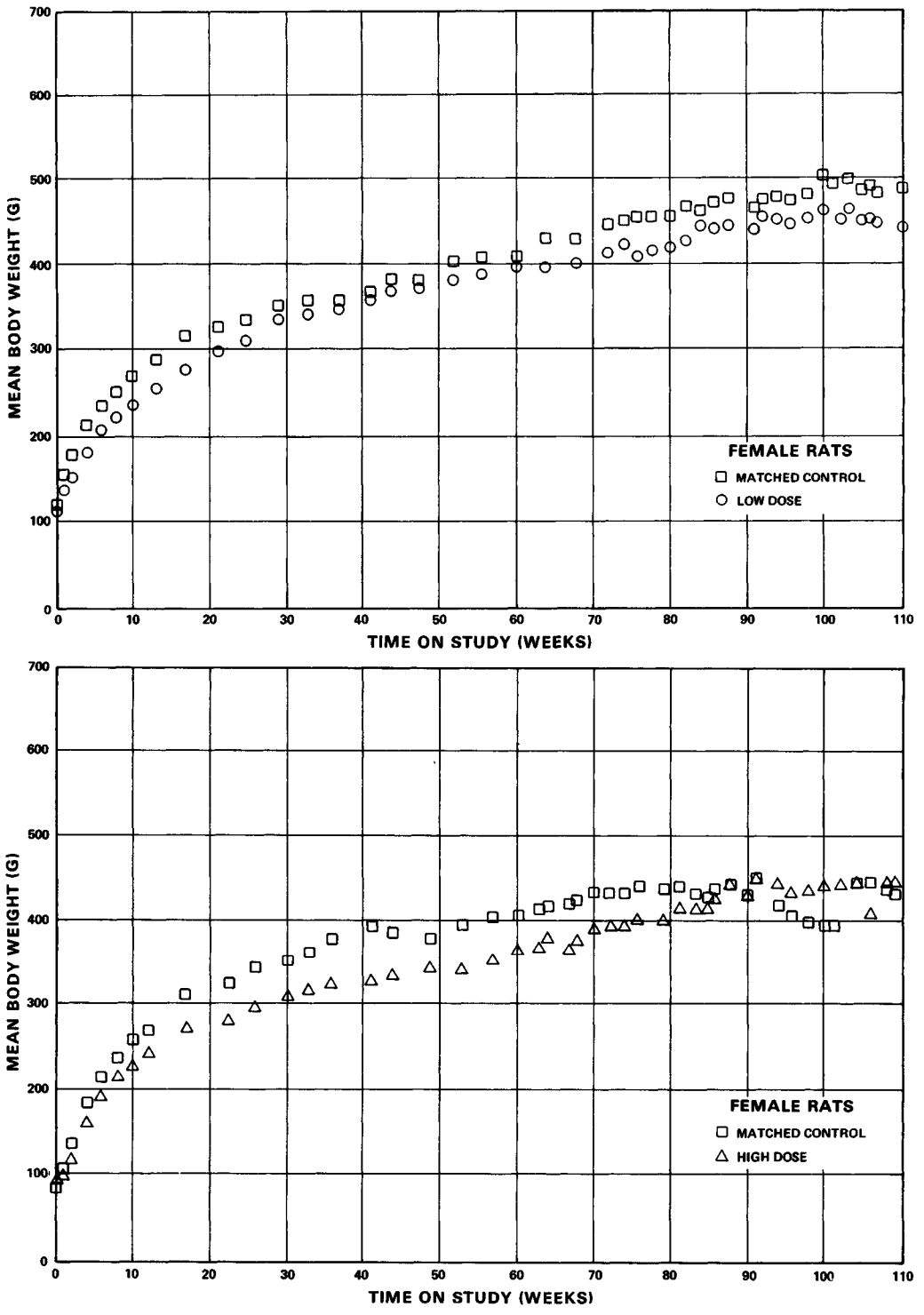


Figure 2. Growth Curves for Female Rats Fed Malathion in the Diet

B. Survival (Rats)

The Kaplan and Meier curves estimating the probabilities of survival for male and female rats fed malathion at the doses used in this bioassay, together with those of the matched controls, are shown in figure 3. Two groups of matched controls, one consisting of 5 animals and the other of 10 animals, were used in this study. In the statistical analyses, these two groups were combined to give matched-control groups of 15 animals for each sex.

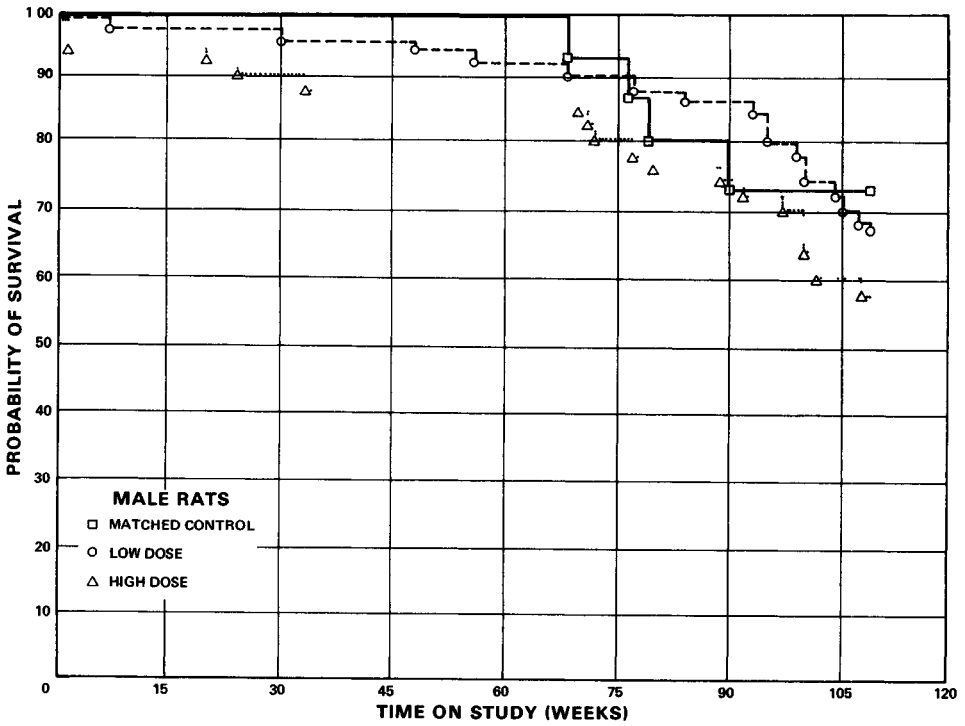
Neither male nor female rats showed a statistically significant dose-related trend in mortality. Fifty-eight percent of the high-dose males and 67% of the high-dose females survived to the end of the study, and survival was higher in the low-dose and control groups.

Sufficient numbers of rats of each sex were at risk for 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.

In general, the majority of the tumors occurred in the dosed



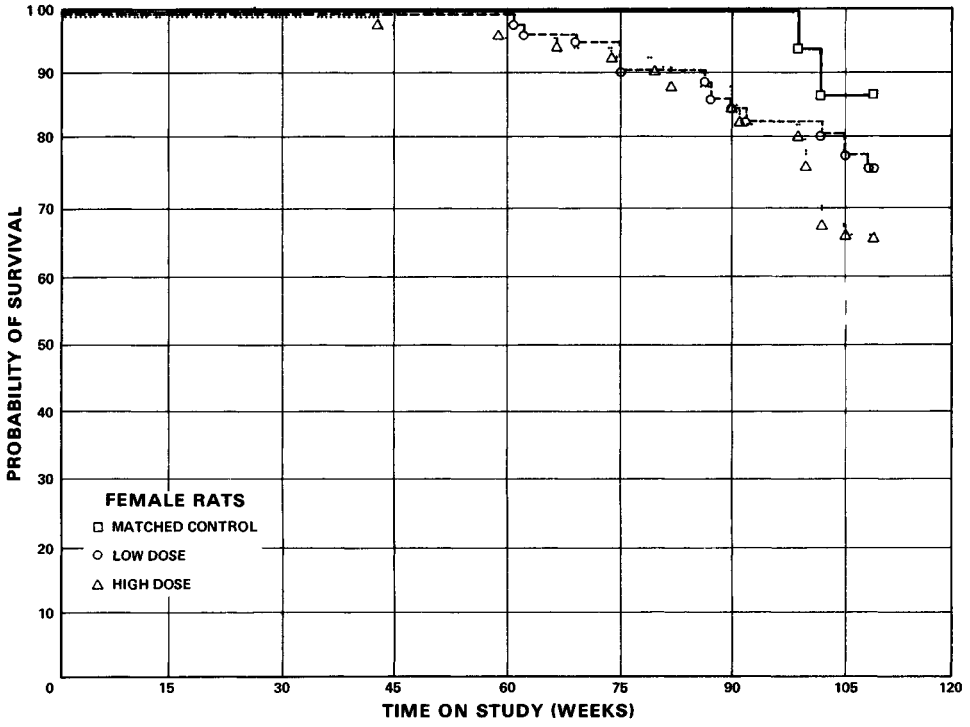


Figure 3. Survival Curves for Rats Fed Malathion in the Diet

animals; however, these tumors were found with a low frequency, except for chromophobe adenomas of the pituitary gland, fibroadenomas of the mammary gland, and follicular-cell carcinomas of the thyroid gland.

There were higher incidences of proliferative lesions of the thyroid gland in the dosed groups than in the matched controls. The incidences of these lesions were as follows:

	MALES			FEMALES		
	Matched Control	Low Dose	High Dose	Matched Control	Low Dose	High Dose
Number of Tissues Examined	(14)	(41)	(47)	(15)	(48)	(49)
Thyroid						
C-cell Hyperplasia	0	1	3	0	5	3
C-cell Adenoma	0	1	3	0	1	2
Follicular-cell Hyperplasia	1	7	8	0	3	0
Follicular-cell Adenoma	1	1	1	0	0	1
Follicular-cell Carcinoma	0	2	6	0	0	3

Follicular-cell carcinomas of the thyroid occurred unilaterally and were nodular in appearance. They varied in size, the largest being 0.9 cm in diameter, while the smallest, 0.2 cm in diameter, was not detected by gross examination. Microscopically, they

were well-differentiated neoplasms made up of densely cellular zones interrupted by less cellular areas containing follicular structures of variable sizes and shapes and containing colloid. Papillary and acinar features occurred essentially with equal frequency. Some lesions appeared to have an incomplete connective tissue capsule through and beyond which neoplastic follicular cells had extended. Invasive growth into preexisting thyroid parenchyma was common, and an occasional example of extension into surrounding musculature was also seen. The cells were somewhat small and cuboidal in outline with pale eosinophilic cytoplasm. Nuclei were relatively large and essentially oval in outline, having well-delineated nuclear membranes and ample, finely stippled nucleoplasm. Mitoses were infrequent. Degenerative changes were evident, but not abundant. Distant metastases were not observed.

Follicular-cell hyperplasia occurred unilaterally, and foci were small in size, thus undetected by gross examination. Microscopically, a single focus, or occasionally two foci, occurred within the thyroid lobe. These foci were unencapsulated and did not seem to be compressing the surrounding thyroid tissue to any great degree. They consisted of several thyroid follicles of variable sizes; many contained normal-appearing colloid. The epithelial wall of these follicles was most commonly cuboidal in

appearance, but epithelial height showed somewhat of an inverse relationship to the diameter of the follicle. Within the larger follicles, thin papillary infoldings in the epithelial wall, suggestive of a colloid involutionary change, were observed. This lesion was thought to be most compatible with a reactive-metabolic change. Microscopically, there was a more or less diffuse increase in the number of interfollicular cells in the thyroid. The cells were pale-staining and polygonal in shape with ill-defined borders. There was a subtle tendency toward nesting. The cytoplasm was somewhat amphophilic and fairly stippled. The nuclei were also pale and uniformly stippled. These cells were positioned around and between thyroid follicles and seemed to encroach upon the follicles, probably causing a reduction in their size. These changes could be seen in one or both lobes, and occasionally in the same lobe with follicular-cell hyperplasia or follicular-cell adenoma.

Numerous nonproliferative lesions, including testicular atrophy and chronic inflammation of the kidney, occurred with low frequency in the control and dosed rats. In addition, cysts of the pituitary gland were seen in 12/43 (28%) low-dose and 10/46 (22%) high-dose males receiving malathion, but the dosed female rats, except for one high-dose female, did not have these lesions of the pituitary gland.

The neoplastic and nonneoplastic lesions that occurred in the rats used in this study appeared in a variety of tissues of most organ systems, and for the most part, the incidences in dosed and control animals were comparable. In a few instances, lesions appeared in dosed animals only, but not in significant numbers. This was particularly true for some of the neoplastic lesions in the endocrine and reproductive systems of the rats. These pathologic changes have commonly been observed in control animals of the same strain in other studies of carcinogenicity and are, therefore, considered to be coincidental. In the judgment of the pathologists, this study shows no evidence of carcinogenicity induced by administration of malathion in the Osborne-Mendel rats at the doses used.

D. Statistical Analyses of Results (Rats)

Tables E1 and E2 in Appendix E contain the statistical analyses of the incidences of those specific primary tumors that occurred in at least two animals in one group and with an incidence of at least 5% in one or more than one group.

The incidences of tumors in the matched and pooled controls in this study do not differ statistically from the incidences seen in data compiled to date from similar studies in the same laboratory.

In male rats, the Cochran-Armitage test results for dose-related trend had levels of significance greater than P = 0.05 for incidences of all tumors listed. Also, the results of the Fisher exact test for the comparison of dosed groups with either group of controls were not statistically significant. There was no statistical evidence, therefore, for carcinogenicity of malathion in the male rats.

In female rats, follicular-cell carcinoma or follicular-cell adenoma of the thyroid appeared in 4/49 (8%) of the high-dose group, but in no animals in either set of controls. The Cochran-Armitage test indicates a significant (P = 0.026) positive linear trend in incidence using pooled controls; however, the results of the Fisher exact test are not significant. The spontaneous rate of this tumor in female Osborne-Mendel rats as observed to date at this laboratory is 6/271 (2.2%). No other tumor in the rats appeared at any site in statistically significant incidences. In each of the 95% confidence intervals, shown in the tables, 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, indicating the theoretical possibility of the induction of tumors in rats from malathion, which could not be detected under the conditions of this test.

IV. RESULTS - MICE

A. Body Weights and Clinical Signs (Mice)

Throughout the study, the mean body weights of the dosed male and female mice were lower than those of their controls (figure 4). The data also indicate dose-related effects of malathion on mean body weight. Fluctuations in the growth curve may be due to mortality; as a size of the group diminishes, the mean body weight may be subject to wide variation.

During the first 6 months of the study, the dosed animals were generally comparable to the controls in appearance and behavior. At week 31, all of the female controls appeared to be hyper-excitable, but this condition did not persist.

During the second year of the study, clinical signs including alopecia, rough and discolored hair coats, poor food consumption, hyperexcitability, and abdominal distention were noted with increasing frequency in dosed animals. A few animals appeared to be hyporeactive, and some had a hunched appearance. At week 71, five high-dose females exhibited generalized body tremors, a condition which persisted until week 79. At week 72, a majority of the high-dose males and females began coughing and sneezing; this condition persisted until termination of the study.

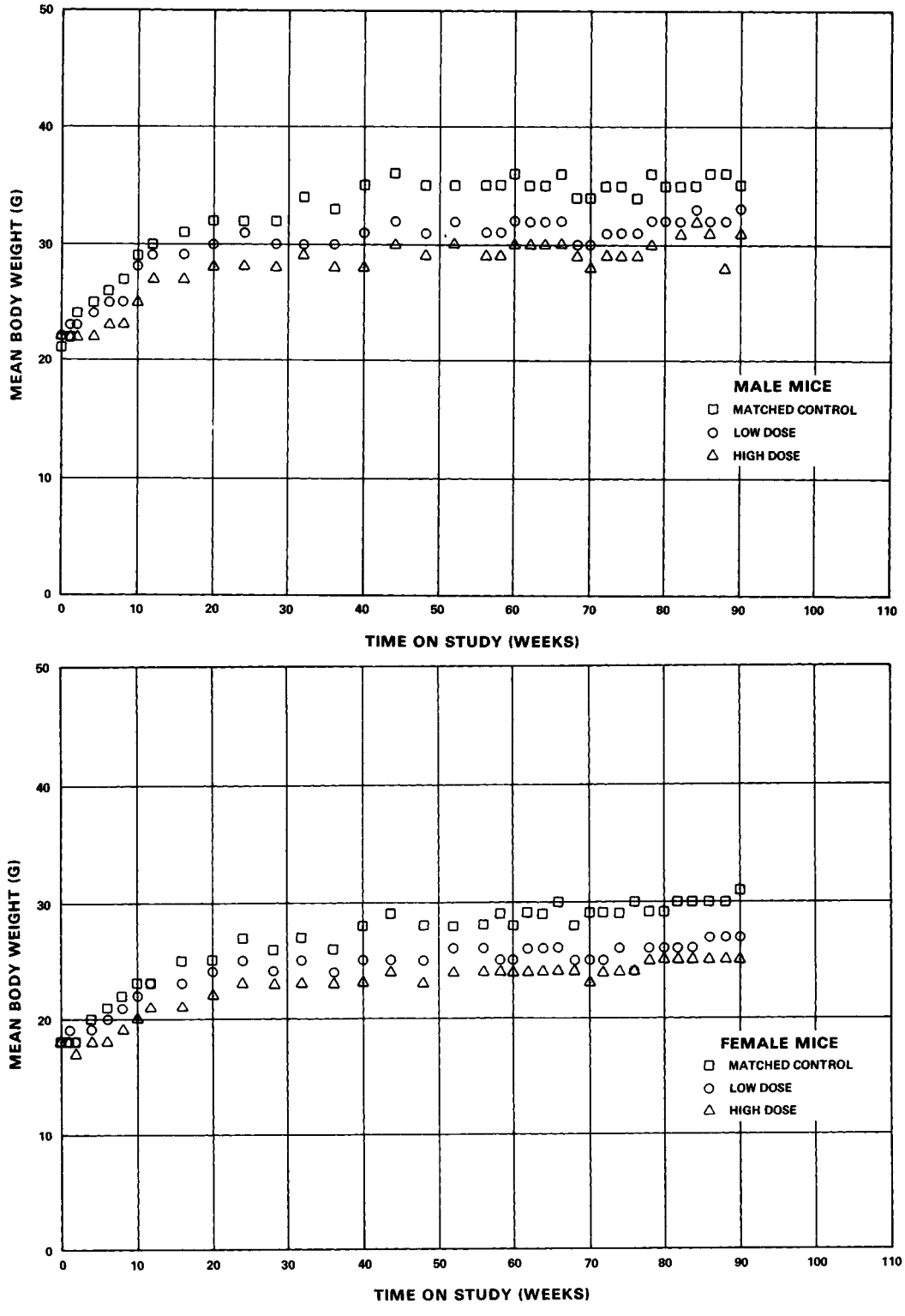


Figure 4. Growth Curves for Mice Fed Malathion in the Diet

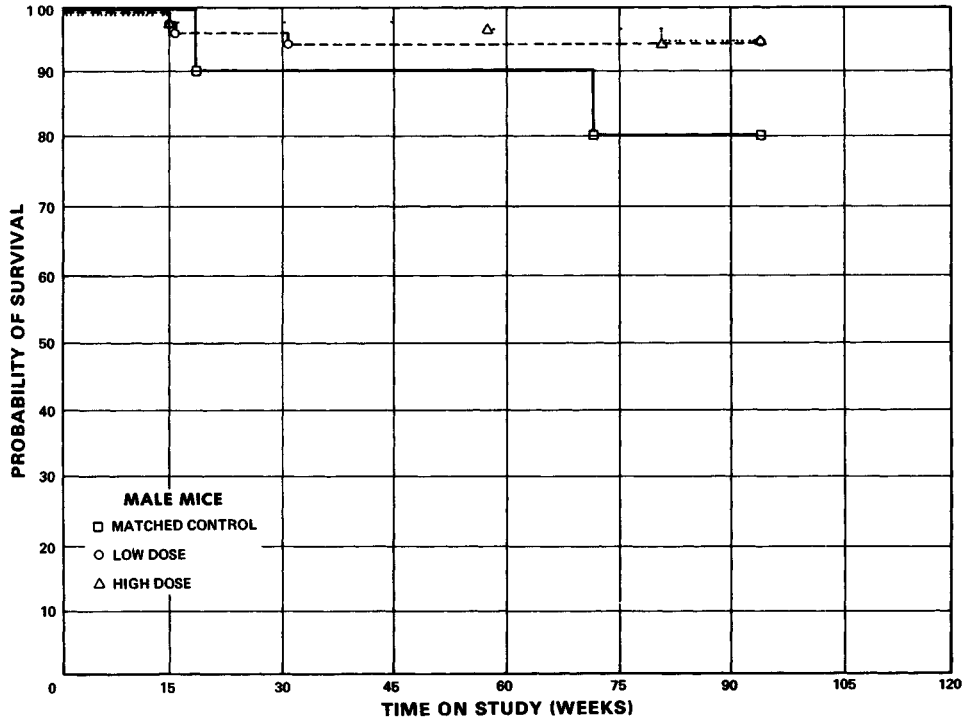
B. Survival (Mice)

The Kaplan and Meier curves estimating the probabilities of survival for male and female mice fed malathion at the doses used in this bioassay, together with those of the matched controls, are shown in figure 5. Neither male nor female mice exhibited a statistically significant dose-related trend in mortality. Ninety-four percent of the high-dose males and 88% of the high-dose females survived to termination of the study. Survival in each sex was higher in the dosed than in the control group. Sufficient numbers of animals of each sex were at risk for 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.

In mice, the only frequently occurring tumor was hepatocellular carcinoma in the males (controls 2/10 [20%], low-dose 7/48 [15%], high-dose 11/49 [22%]); none were found in female mice. The hepatocellular carcinomas varied in microscopic tinctorial qualities, architectural patterns, and cytologic morphology. Some of these lesions, particularly the smaller ones, were observed, not at necropsy, but only after the liver had been



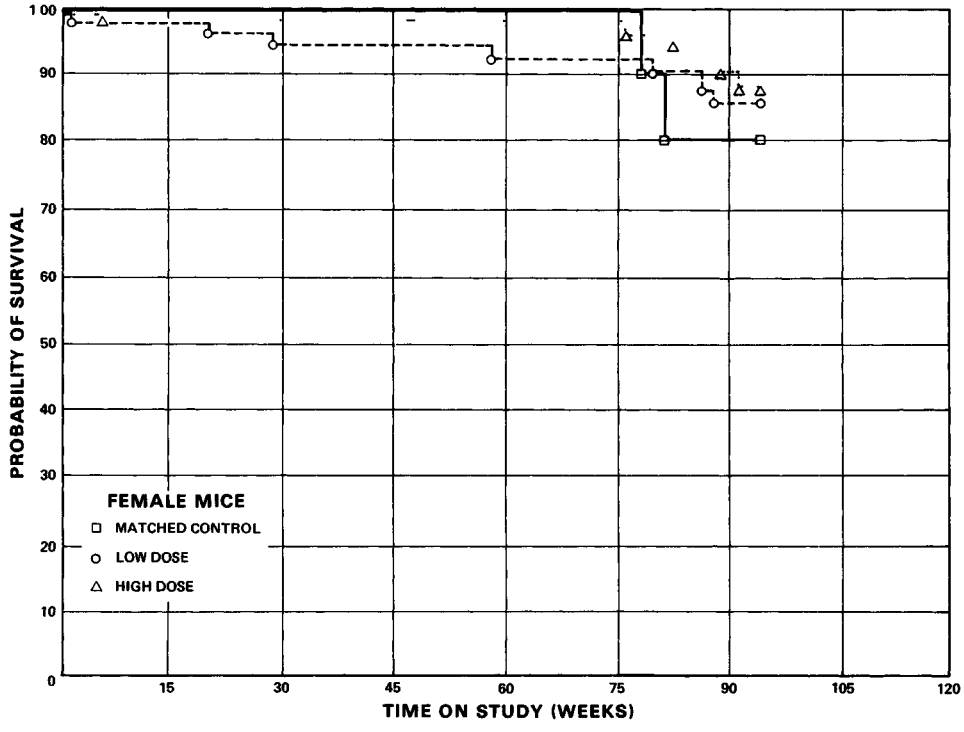


Figure 5. Survival Curves for Mice Fed Malathion in the Diet

fixed. Grossly, the neoplasms varied in size from microscopic to 2-3 cm in diameter, and were usually round and well limited. Some had a multinodular appearance, and their boundaries were not conspicuous. The tumors were located either immediately under the capsule and bulged on the visceral surface or were deep within the parenchyma. In the majority of cases, a single tumor was found, but multifocal lesions were also observed.

Microscopically, the tumors were clearly discernible, because of changes in the architectural and/or tinctorial characteristics that distinguish the neoplastic from the normal tissues. In addition, the surrounding normal liver plates were compressed and narrowed, due to expansion of the tumor. No capsule formation was evident. The neoplastic hepatocytes were variable in size and shape, but often resembled normal liver cells. Anaplastic features were common and served to differentiate malignant and benign neoplastic lesions of the liver. As mentioned previously, the liver cells in the tumors were arranged in different patterns, some of which closely resembled the normal architecture, while others exhibited disorganized or disoriented liver plates. Degenerative changes were often associated with the hepatocellular carcinomas. No metastases to other organs were observed.

Neoplastic nodules of the liver occurred in six high-dose male and two high-dose female mice. These nodules consisted of one or

several small, well-circumscribed nodules located within the hepatic substance. Some of these lesions had gross and cytologic features similar to those of the hepatocellular carcinomas, but size, degree of anaplasia, and lack of invasiveness were the criteria used to distinguish them from hepatocellular carcinomas. Nevertheless, a diagnosis of borderline malignancy was given to a number of the neoplastic nodules. These lesions are considered by the original pathologists to be comparable to neoplastic nodules in rats as described by Squire and Levitt (1975).

A low incidence of miscellaneous degenerative, proliferative, and inflammatory lesions was distributed among the males and females of the control and dosed groups. An exception to this was cystic endometrial hyperplasia, found in 1/9 (11%) control, 12/47 (26%) low-dose, and 10/42 (24%) high-dose female mice.

The neoplastic and nonneoplastic lesions that occurred in the mice used in this study appeared in a variety of tissues of most organ systems, and for the most part, with similar incidences for dosed and control animals. In a few instances, lesions occurred only in animals of the dosed groups, but not in significant numbers. These pathologic changes have been observed in control animals in other studies of carcinogenicity and are, therefore, not considered dose related. In the judgment of the pathologists, this study shows no evidence of carcinogenicity

induced by the administration of malathion in B6C3F1 mice at the doses used.

D. Statistical Analyses of Results (Mice)

Tables F1 and F2 of Appendix F contain the statistical analyses of the incidences of those specific primary tumors that occurred in at least two animals in one group and with an incidence of at least 5% in one or more than one group.

In male mice, the incidence of hepatocellular carcinoma combined with that of neoplastic nodules showed a significant linear trend when either the matched controls ($P = 0.041$) or the pooled controls ($P = 0.019$) were used. The Fisher exact test for the comparison between the results of the high-dose (17/49, 35%) and pooled-control groups had a probability level of $P = 0.031$, but this probability of 0.031 is above the 0.025 level required by the application of the Bonferroni criterion. When time-adjusted analysis is performed, eliminating those male mice that died before 52 weeks on study, the following incidences resulted: matched controls, 2/9 (22%); pooled control, 8/48 (17%); low-dose, 7/47 (15%); high-dose, 17/49 (35%). Neither the Fisher exact tests nor the Cochran-Armitage test of these time-adjusted incidences are significant ($P > 0.05$) when the matched controls are used. It should be observed that when the incidences of

hepatocellular carcinoma and neoplastic nodules were analyzed separately, none of the Fisher exact test results were significant. The results of all the similar bioassays from this laboratory for which data are available for the B6C3F1 mouse show an incidence of spontaneous liver tumors of 19% of 285 male mice; this is close to the incidence (16%) seen in the male pooled-control group in the present bioassay. In female mice, there were no statistically significant incidences of any tumor in the dosed groups when compared with those of either set of controls. In each of the 95% confidence intervals, shown in the tables, 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, indicating the theoretical possibility of the induction of tumors by this chemical, which could not be detected under the conditions of this test.

V. DISCUSSION

Malathion is a member of the organophosphorus class of pesticides whose predominant mode of inducing toxicity is by inhibition of cholinesterase. Cholinesterase activity in the cerebral cortex, erythrocytes, and plasma of rats is reduced when the animals are fed diets containing malathion (Desi et al., 1976) or when the chemical is administered by oral intubation (Mendoza, 1976).

Mean body weights for females of either species were lower for the dosed groups than for the matched controls. Other clinical signs also appeared with greater frequency in the dosed animals than in the controls. In rats, these included rough hair coats, pale mucous membranes, dermatitis, ataxia, alopecia, hematuria, and vaginal bleeding; in mice, the signs included rough and discolored hair coats and abdominal distention. Mortality in either rats or mice was not significantly related to the administration of malathion. Sufficient numbers of animals were at risk in all groups of rats and mice to termination of the study for the development of late-appearing tumors.

In female rats, three follicular-cell carcinomas and one follicular-cell adenoma of the thyroid occurred in the high-dose group, and three follicular-cell hyperplasias occurred in the low-dose group. The incidence of these tumors showed a statistically

significant (P = 0.026) dose-related trend; however, the results of the Fisher exact test for direct comparison between the dosed and control groups were not significant. More dosed males than dosed females had either tumors or hyperplasia of the follicular cells of the thyroid; however, because of the higher incidence of tumors among the male controls, the results of none of the statistical tests were significant. These thyroid tumors were not considered to be associated with the administration of malathion.

In male mice, hepatocellular carcinoma occurred at the following incidences: matched controls 2/10, pooled controls 5/49, low-dose 7/48, high-dose 11/49. In addition, neoplastic nodules occurred in 3/49 pooled-control and 6/49 high-dose animals. When the combined incidence in the dosed animals was compared with that of the pooled controls, the dose-related trend was P = 0.019 and the direct comparison of the high-dose group with the control-group was P = 0.031. Thus, none of the direct comparisons of dosed groups with controls were significant using the Bonferroni criteria. In addition, the historical controls for this laboratory had several control groups with incidences of 35-40% hepatocellular carcinomas, rates which are comparable with the incidence of this tumor in dosed male mice in the present study. Thus, these liver tumors are not considered to be

associated with the administration of malathion. The incidences of liver tumors in dosed females were not statistically significant when compared with that in control animals.

In previous studies of the long-term toxicity of malathion, Hazleton and Holland (1953) fed groups of rats 100, 1,000, or 5,000 ppm of either 65, 90, or 99% preparations of malathion for 2-year periods. No lesions associated with the administration of malathion were found by microscopic examination of representative tissues. The highest dose, 5,000 ppm, approximates the time-weighted average low dose used in the present bioassay. The only evidence of the possible induction of tumors from the administration of malathion in the literature is that of the inconclusive studies of Okey (1972), in which Sprague-Dawley rats fed a diet containing 250 ppm malathion showed a higher incidence and shorter induction time of tumors after the administration of dimethylbenzanthracene (1.31 $\pm$ 0.18 tumors/rat) than controls administered dimethylbenzanthracene alone (1.07 $\pm$ 0.15 tumors/rat).

It is concluded that under the conditions of this bioassay, there was no clear evidence of the association of the tumor incidence with the administration of malathion to Osborne-Mendel rats or B6C3F1 mice.

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APPENDIX A

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN RATS FED MALATHION IN THE DIET

TABLE A1.

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS FED MALATHION IN THE DIET

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	5	10	50	50
ANIMALS NECROPSIED	5	10	48	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	5	10	48	50
INTEGUMENTARY SYSTEM				
*SKIN	(5)	(10)	(48)	(50)
BASAL-CELL TUMOR			1 (2%)	
FIBROUS HISTIOCYTOMA			1 (2%)	1 (2%)
FIBROUS HISTIOCYTOMA, MALIGNANT				1 (2%)
RESPIRATORY SYSTEM				
#LUNG	(5)	(10)	(48)	(48)
SQUAMOUS CELL CARCINOMA, METASTA				1 (2%)
OSTEOSARCOMA, METASTATIC			1 (2%)	
HEMATOPOIETIC SYSTEM				
#SPLEEN	(5)	(10)	(47)	(46)
HEMANGIOSARCOMA			1 (2%)	
#THYMUS				(1)
SARCOMA, NOS				1 (100%)
CIRCULATORY SYSTEM				
NONE				
DIGESTIVE SYSTEM				
#SALIVARY GLAND	(5)	(10)	(48)	(47)
FIBROMA	1 (20%)			
#LIVER	(5)	(9)	(47)	(50)
NEOPLASTIC NODULE			1 (2%)	1 (2%)

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

TABLE A1. MALE RATS: NEOPLASMS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
HEPATOCELLULAR CARCINOMA			1 (2%)	
SARCOMA, NOS				1 (2%)
URINARY SYSTEM				
#KIDNEY	(5)	(10)	(48)	(50)
MIXED TUMOR, MALIGNANT			2 (4%)	1 (2%)
HAMARTOMA				1 (2%)
ENDOCRINE SYSTEM				
#PITUITARY	(4)	(6)	(43)	(46)
CARCINOMA,NOS			2 (5%)	1 (2%)
ADENOMA, NOS			1 (2%)	
CHROMOPHOBE ADENOMA			6 (14%)	3 (7%)
#ADRENAL	(5)	(10)	(46)	(50)
CARCINOMA,NOS			1 (2%)	
CORTICAL ADENOMA			2 (4%)	
PHEOCHROMOCYTOMA				3 (6%)
#THYROID	(5)	(9)	(41)	(47)
FOLLICULAR-CELL ADENOMA	1 (20%)		1 (2%)	1 (2%)
FOLLICULAR-CELL CARCINOMA			2 (5%)	6 (13%)
C-CELL ADENOMA			1 (2%)	3 (6%)
#PANCREATIC ISLETS	(5)	(10)	(45)	(49)
ISLET-CELL ADENOMA			1 (2%)	3 (6%)
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(5)	(10)	(48)	(50)
FIBROMA		1 (10%)		
NERVOUS SYSTEM				
#BRAIN	(5)	(10)	(48)	(48)
MENINGIOMA				1 (2%)
#CEREBELLUM	(5)	(10)	(48)	(48)
GRANULAR-CELL TUMOR, BENIGN				1 (2%)
*TRIGEMINAL NERVE	(5)	(10)	(48)	(50)
SQUAMOUS CELL CARCINOMA				1 (2%)

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

TABLE A1. MALE RATS: NEOPLASMS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
SPECIAL SENSE ORGANS				
NONE				
MUSCULOSKELETAL SYSTEM				
*HUMERUS	(5)	(10)	(48)	(50)
OSTEOSARCOMA			1 (2%)	
BODY CAVITIES				
*ABDOMINAL CAVITY	(5)	(10)	(48)	(50)
LIPOMA		1 (10%)		
ALL OTHER SYSTEMS				
*MULTIPLE ORGANS	(5)	(10)	(48)	(50)
FIBROUS HISTIOCYTOMA, MALIGNANT			1 (2%)	
ANIMAL DISPOSITION SUMMARY				
ANIMALS INITIALLY IN STUDY	5	10	50	50
NATURAL DEATH@			7	9
MORIBUND SACRIFICE	5		10	12
SCHEDULED SACRIFICE				
ACCIDENTALLY KILLED				
TERMINAL SACRIFICE		10	33	29
ANIMAL MISSING				

@ INCLUDES AUTOLYZED ANIMALS

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

TABLE A1. MALE RATS: NEOPLASMS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
TUMOR SUMMARY				
TOTAL ANIMALS WITH PRIMARY TUMORS*	2	2	22	23
TOTAL PRIMARY TUMORS	2	2	26	30
TOTAL ANIMALS WITH BENIGN TUMORS	2	2	14	13
TOTAL BENIGN TUMORS	2	2	14	16
TOTAL ANIMALS WITH MALIGNANT TUMORS			10	12
TOTAL MALIGNANT TUMORS			11	13
TOTAL ANIMALS WITH SECONDARY TUMORS#			1	1
TOTAL SECONDARY TUMORS			1	1
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 FED MALATHION IN THE DIET

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	5	10	50	50
ANIMALS NECROPSIED	5	10	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	5	10	50	50
INTEGUMENTARY SYSTEM				
*SUBCUT TISSUE	(5)	(10)	(50)	(50)
FIBROSARCOMA			1 (2%)	1 (2%)
RESPIRATORY SYSTEM				
*NASAL CAVITY	(5)	(10)	(50)	(50)
CARCINOMA,NOS				1 (2%)
#LUNG	(5)	(10)	(50)	(50)
ALVEOLAR/BRONCHIOLAR ADENOMA			1 (2%)	
FIBROSARCOMA, METASTATIC			1 (2%)	1 (2%)
HEMATOPOIETIC SYSTEM				
#SPLEEN	(5)	(10)	(49)	(49)
HEMANGIOMA			1 (2%)	1 (2%)
CIRCULATORY SYSTEM				
NONE				
DIGESTIVE SYSTEM				
#LIVER	(5)	(10)	(50)	(49)
NEOPLASTIC NODULE				2 (4%)
HEPATOCELLULAR CARCINOMA				1 (2%)
HEMANGIOMA			1 (2%)	
URINARY SYSTEM				
#KIDNEY	(5)	(10)	(50)	(50)
MIXED TUMOR, MALIGNANT		1 (10%)	1 (2%)	

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

TABLE A2. FEMALE RATS: NEOPLASMS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM				
#PITUITARY	(5)	(10)	(42)	(45)
CARCINOMA,NOS			2 (5%)	
ADENOMA, NOS				1 (2%)
CHROMOPHOBE ADENOMA		3 (30%)	8 (19%)	7 (16%)
CHROMOPHOBE CARCINOMA				1 (2%)
#ADRENAL	(5)	(10)	(49)	(48)
CORTICAL ADENOMA			3 (6%)	
PHEOCHROMOCYTOMA		1 (10%)		
#THYROID	(5)	(10)	(48)	(49)
FOLLICULAR-CELL ADENOMA				1 (2%)
FOLLICULAR-CELL CARCINOMA				3 (6%)
C-CELL ADENOMA			1 (2%)	2 (4%)
#PANCREATIC ISLETS	(5)	(10)	(50)	(50)
ISLET-CELL ADENOMA			2 (4%)	
ISLET-CELL CARCINOMA				1 (2%)
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(5)	(10)	(50)	(50)
ADENOMA, NOS			1 (2%)	1 (2%)
ADENOCARCINOMA, NOS				1 (2%)
FIBROMA			2 (4%)	1 (2%)
FIBROADENOMA			6 (12%)	7 (14%)
#UTERUS	(5)	(10)	(48)	(46)
LEIOMYOSARCOMA				1 (2%)
ENDOMETRIAL STROMAL POLYP		2 (20%)	1 (2%)	1 (2%)
NERVOUS SYSTEM				
#BRAIN	(5)	(10)	(50)	(49)
CARCINOMA, NOS, METASTATIC				1 (2%)
HAMARTOMA			1 (2%)	
GRANULAR-CELL TUMOR, BENIGN			1 (2%)	1 (2%)
GLIOMA, NOS				1 (2%)
SPECIAL SENSE ORGANS				
NONE				

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

TABLE A2. FEMALE RATS: NEOPLASMS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
*MULTIPLE ORGANS	(5)	(10)	(50)	(50)
FIBROUS HISTIOCYTOMA, MALIGNANT				1 (2%)
ANIMAL DISPOSITION SUMMARY				
ANIMALS INITIALLY IN STUDY	5	10	50	50
NATURAL DEATH@			2	1
MORIBUND SACRIFICE	1	1	10	18
SCHEDULED SACRIFICE				
ACCIDENTALLY KILLED				
TERMINAL SACRIFICE	4	9	38	31
ANIMAL MISSING				

@ INCLUDES AUTOLYZED ANIMALS

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

TABLE A2. FEMALE RATS: NEOPLASMS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
TUMOR SUMMARY				
TOTAL ANIMALS WITH PRIMARY TUMORS*		6	23	28
TOTAL PRIMARY TUMORS		7	33	37
TOTAL ANIMALS WITH BENIGN TUMORS		6	20	21
TOTAL BENIGN TUMORS		6	29	23
TOTAL ANIMALS WITH MALIGNANT TUMORS		1	4	12
TOTAL MALIGNANT TUMORS		1	4	12
TOTAL ANIMALS WITH SECONDARY TUMORS#			1	2
TOTAL SECONDARY TUMORS			1	2
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT				2
TOTAL UNCERTAIN TUMORS				2
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 FED MALATHION IN THE DIET

TABLE B1.

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE FED MALATHION IN THE DIET

	CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	10	50	50
ANIMALS NECROPSIED	10	48	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY	10	48	49
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG	(9)	(48)	(49)
ALVEOLAR/BRONCHIOLAR ADENOMA		1 (2%)	1 (2%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(10)	(48)	(49)
LYMPHOCYTIC LEUKEMIA	1 (10%)		
#BONE MARROW	(10)	(48)	(46)
LEUKEMIA,NOS			1 (2%)
#LIVER	(10)	(48)	(49)
LEUKEMIA,NOS			1 (2%)
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(9)	(48)	(47)
ADENOCARCINOMA, NOS			1 (2%)
#LIVER	(10)	(48)	(49)
NEOPLASTIC NODULE			6 (12%)
HEPATOCELLULAR CARCINOMA	2 (20%)	7 (15%)	11 (22%)

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

TABLE B1. MALE MICE: NEOPLASMS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
HEMANGIOMA		1 (2%)	
URINARY SYSTEM			
NONE			
ENDOCRINE SYSTEM			
NONE			
REPRODUCTIVE SYSTEM			
NONE			
NERVOUS SYSTEM			
NONE			
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. MALE MICE: NEOPLASMS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	10	50	50
NATURAL DEATH@	1	3	1
MORIBUND SACRIFICE	1		2
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	8	47	47
ANIMAL MISSING			
@ INCLUDES AUTOLYZED ANIMALS			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	3	9	19
TOTAL PRIMARY TUMORS	3	9	21
TOTAL ANIMALS WITH BENIGN TUMORS		2	1
TOTAL BENIGN TUMORS		2	1
TOTAL ANIMALS WITH MALIGNANT TUMORS	3	7	12
TOTAL MALIGNANT TUMORS	3	7	14
TOTAL ANIMALS WITH SECONDARY TUMORS#			
TOTAL SECONDARY TUMORS			
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT			6
TOTAL UNCERTAIN TUMORS			6
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 FED MALATHION IN THE DIET

	CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	10	50	50
ANIMALS NECROPSIED	10	49	48
ANIMALS EXAMINED HISTOPATHOLOGICALLY	10	49	47
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
NONE			
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(10)	(49)	(48)
MALIGNANT LYMPHOMA, NOS		1 (2%)	
LEUKEMIA,NOS			1 (2%)
GRANULOCYTIC LEUKEMIA	1 (10%)		
#SPLEEN	(10)	(48)	(46)
HEMANGIOMA			1 (2%)
#LIVER	(10)	(49)	(47)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE			1 (2%)
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#LIVER	(10)	(49)	(47)
NEOPLASTIC NODULE			2 (4%)
URINARY SYSTEM			
NONE			

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

TABLE B2. FEMALE MICE: NEOPLASMS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM			
#PITUITARY	(7)	(39)	(39)
CHROMOPHOBE ADENOMA		1 (3%)	
#THYROID	(9)	(38)	(40)
FOLLICULAR-CELL ADENOMA		1 (3%)	
REPRODUCTIVE SYSTEM			
#UTERUS	(9)	(47)	(42)
LEIOMYOMA		1 (2%)	
#OVARY	(10)	(48)	(45)
TERATOMA, BENIGN		1 (2%)	
NERVOUS SYSTEM			
NONE			
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 B2. FEMALE MICE: NEOPLASMS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	10	50	50
NATURAL DEATH@	1	2	4
MORIBUND SACRIFICE	1	5	2
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	8	43	44
ANIMAL MISSING			
@ INCLUDES AUTOLYZED ANIMALS			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	1	5	5
TOTAL PRIMARY TUMORS	1	5	5
TOTAL ANIMALS WITH BENIGN TUMORS		4	1
TOTAL BENIGN TUMORS		4	1
TOTAL ANIMALS WITH MALIGNANT TUMORS	1	1	2
TOTAL MALIGNANT TUMORS	1	1	2
TOTAL ANIMALS WITH SECONDARY TUMORS#			
TOTAL SECONDARY TUMORS			
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT			2
TOTAL UNCERTAIN TUMORS			2
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 FED MALATHION IN THE DIET

TABLE C1.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS FED MALATHION IN THE DIET

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	5	10	50	50
ANIMALS NECROPSIED	5	10	48	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	5	10	48	50
INTEGUMENTARY SYSTEM				
*SKIN	(5)	(10)	(48)	(50)
INFLAMMATION, GRANULOMATOUS			1 (2%)	
GRANULATION, TISSUE			2 (4%)	
RESPIRATORY SYSTEM				
#LUNG	(5)	(10)	(48)	(48)
ATELECTASIS			1 (2%)	
CONGESTION, NOS			1 (2%)	
EDEMA, NOS			1 (2%)	
INFLAMMATION, CHRONIC FOCAL			2 (4%)	1 (2%)
#LUNG/ALVEOLI	(5)	(10)	(48)	(48)
CALCIFICATION, METASTATIC				1 (2%)
HEMATOPOIETIC SYSTEM				
#SPLEEN	(5)	(10)	(47)	(46)
FIBROSIS				2 (4%)
FIBROSIS, FOCAL				2 (4%)
#LYMPH NODE	(5)	(10)	(44)	(42)
INFLAMMATION, CHRONIC		1 (10%)		
CIRCULATORY SYSTEM				
#HEART	(5)	(10)	(47)	(50)
THROMBOSIS, NOS	1 (20%)			
THROMBUS, ORGANIZED			1 (2%)	
ARTERIOSCLEROSIS, NOS				1 (2%)
#MYOCARDIUM	(5)	(10)	(47)	(50)
INFLAMMATION, INTERSTITIAL			1 (2%)	

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

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
FIBROSIS			1 (2%)	
FIBROSIS, FOCAL			1 (2%)	2 (4%)
#ENDOCARDIUM	(5)	(10)	(47)	(50)
FIBROSIS		1 (10%)		3 (6%)
FIBROSIS, FOCAL				1 (2%)
*AORTA	(5)	(10)	(48)	(50)
ARTERIOSCLEROSIS, NOS				1 (2%)
MEDIAL CALCIFICATION			1 (2%)	
DIGESTIVE SYSTEM				
#LIVER	(5)	(9)	(47)	(50)
INFLAMMATION, FOCAL GRANULOMATOU	1 (20%)			
GRANULOMA, FOREIGN BODY			1 (2%)	
DEGENERATION, BALLOONING			2 (4%)	
DEGENERATION PARENCHYMATOUS			1 (2%)	4 (8%)
METAMORPHOSIS FATTY	1 (20%)		2 (4%)	2 (4%)
FOCAL CELLULAR CHANGE				2 (4%)
ANGIECTASIS			1 (2%)	
#HEPATIC CAPSULE	(5)	(9)	(47)	(50)
ANGIECTASIS				1 (2%)
*BILE DUCT	(5)	(10)	(48)	(50)
INFLAMMATION, CHRONIC			1 (2%)	
HYPERPLASIA, NOS		1 (10%)	1 (2%)	1 (2%)
HYPERPLASIA, FOCAL			1 (2%)	1 (2%)
HYPERPLASIA, DIFFUSE				1 (2%)
#PANCREAS	(5)	(10)	(45)	(49)
INFLAMMATION, CHRONIC				1 (2%)
PERIARTERITIS			3 (7%)	
#PANCREATIC ACINUS	(5)	(10)	(45)	(49)
ATROPHY, NOS			1 (2%)	1 (2%)
#STOMACH	(5)	(10)	(48)	(47)
CALCIFICATION, METASTATIC	1 (20%)		2 (4%)	3 (6%)
URINARY SYSTEM				
#KIDNEY	(5)	(10)	(48)	(50)
INFLAMMATION, CHRONIC	4 (80%)	4 (40%)	33 (69%)	33 (66%)

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

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
#URINARY BLADDER	(5)	(10)	(45)	(42)
INFLAMMATION, ACUTE DIFFUSE				1 (2%)
INFLAMMATION, CHRONIC			1 (2%)	
INFLAMMATION, CHRONIC FOCAL			1 (2%)	
ENDOCRINE SYSTEM				
#PITUITARY	(4)	(6)	(43)	(46)
CYST, NOS			12 (28%)	10 (22%)
HYPERPLASIA, NOS			1 (2%)	2 (4%)
HYPERPLASIA, FOCAL			1 (2%)	2 (4%)
#ADRENAL	(5)	(10)	(46)	(50)
DEGENERATION, CYSTIC			1 (2%)	
#ADRENAL CORTEX	(5)	(10)	(46)	(50)
METAMORPHOSIS FATTY			1 (2%)	
HYPERPLASIA, FOCAL			3 (7%)	2 (4%)
#THYROID	(5)	(9)	(41)	(47)
CYSTIC FOLLICLES		1 (11%)		3 (6%)
FOLLICULAR CYST, NOS			1 (2%)	
MULTIPLE CYSTS				1 (2%)
ATROPHY, NOS				1 (2%)
HYPERPLASIA, C-CELL			1 (2%)	3 (6%)
HYPERPLASIA, FOLLICULAR-CELL		1 (11%)	7 (17%)	8 (17%)
METAPLASIA, SQUAMOUS				1 (2%)
#THYROID FOLLICLE	(5)	(9)	(41)	(47)
ATROPHY, NOS				1 (2%)
#PARATHYROID	(3)	(3)	(24)	(26)
HYPERPLASIA, NOS	2 (67%)		2 (8%)	1 (4%)
HYPERPLASIA, DIFFUSE				1 (4%)
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(5)	(10)	(48)	(50)
CYST, NOS		1 (10%)		
HYPERPLASIA, NOS	1 (20%)			
DYSPLASIA, NOS			1 (2%)	
#PROSTATE	(5)	(10)	(45)	(48)
INFLAMMATION, SUPPURATIVE	1 (20%)		1 (2%)	

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

TABLE C1. MALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
INFLAMMATION, ACUTE SUPPURATIVE		1 (10%)		1 (2%)
INFLAMMATION, ACUTE NECROTIZING				1 (2%)
INFLAMMATION ACUTE AND CHRONIC		1 (10%)	2 (4%)	
#TESTIS	(5)	(9)	(45)	(49)
PERIARTERITIS			2 (4%)	
ATROPHY, NOS	1 (20%)	1 (11%)	11 (24%)	4 (8%)
HYPERPLASIA, INTERSTITIAL CELL				1 (2%)
NERVOUS SYSTEM				
#BRAIN/MENINGES	(5)	(10)	(48)	(48)
MULTIPLE CYSTS				1 (2%)
SPECIAL SENSE ORGANS				
NONE				
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
*MESENTERY	(5)	(10)	(48)	(50)
PERIARTERITIS			4 (8%)	
ALL OTHER SYSTEMS				
NONE				
SPECIAL MORPHOLOGY SUMMARY				
NO LESION REPORTED		4	3	7
AUTOLYSIS/NO NECROPSY			2	

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

TABLE C2.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS FED MALATHION IN THE DIET

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	5	10	50	50
ANIMALS NECROPSIED	5	10	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	5	10	50	50
INTEGUMENTARY SYSTEM				
*SKIN	(5)	(10)	(50)	(50)
ULCER, NOS			1 (2%)	
RESPIRATORY SYSTEM				
#LUNG	(5)	(10)	(50)	(50)
INFLAMMATION, FOCAL				1 (2%)
INFLAMMATION, ACUTE HEMORRHAGIC			1 (2%)	
HEMATOPOIETIC SYSTEM				
#SPLEEN	(5)	(10)	(49)	(49)
INFLAMMATION, CHRONIC				1 (2%)
HYPERPLASIA, RETICULUM CELL				1 (2%)
MYELOID METAPLASIA		1 (10%)		
#PANCREATIC L.NODE	(5)	(9)	(42)	(43)
INFLAMMATION, NOS		1 (11%)		
CIRCULATORY SYSTEM				
#MYOCARDIUM	(5)	(10)	(50)	(50)
INFLAMMATION, CHRONIC FOCAL				1 (2%)
FIBROSIS, FOCAL			1 (2%)	
#ENDOCARDIUM	(5)	(10)	(50)	(50)
FIBROSIS			1 (2%)	
DIGESTIVE SYSTEM				
#LIVER	(5)	(10)	(50)	(49)
CONGESTION, NOS	1 (20%)			

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

TABLE C2. FEMALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
DEGENERATION PARENCHYMATOUS				2 (4%)
NECROSIS, NOS				1 (2%)
NECROSIS, FOCAL			1 (2%)	
METAMORPHOSIS FATTY	1 (20%)	2 (20%)	2 (4%)	8 (16%)
FOCAL CELLULAR CHANGE			1 (2%)	2 (4%)
ANGIECTASIS	1 (20%)		2 (4%)	8 (16%)
*BILE DUCT	(5)	(10)	(50)	(50)
INFLAMMATION, NOS			1 (2%)	2 (4%)
INFLAMMATION, ACUTE				1 (2%)
INFLAMMATION, CHRONIC				1 (2%)
HYPERPLASIA, NOS			1 (2%)	4 (8%)
HYPERPLASIA, FOCAL			2 (4%)	
#PANCREATIC ACINUS	(5)	(10)	(50)	(50)
ATROPHY, FOCAL			1 (2%)	1 (2%)
#STOMACH	(5)	(10)	(48)	(49)
EROSION			1 (2%)	
URINARY SYSTEM				
#KIDNEY	(5)	(10)	(50)	(50)
GLOMERULONEPHRITIS, NOS				1 (2%)
INFLAMMATION, CHRONIC	2 (40%)		3 (6%)	11 (22%)
INFARCT, NOS				1 (2%)
#KIDNEY/CORTEX	(5)	(10)	(50)	(50)
CYST, NOS			1 (2%)	
#KIDNEY/TUBULE	(5)	(10)	(50)	(50)
DEGENERATION, NOS				1 (2%)
ENDOCRINE SYSTEM				
#PITUITARY	(5)	(10)	(42)	(45)
CYST, NOS				1 (2%)
CONGESTION, NOS				1 (2%)
HEMORRHAGE			1 (2%)	
DEGENERATION, CYSTIC				1 (2%)
HYPERPLASIA, NOS			1 (2%)	
HYPERPLASIA, FOCAL			3 (7%)	
ANGIECTASIS				3 (7%)
#ADRENAL	(5)	(10)	(49)	(48)
DEGENERATION, CYSTIC			2 (4%)	

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

TABLE C2. FEMALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
NECROSIS, NOS			1 (2%)	
METAMORPHOSIS FATTY				1 (2%)
ANGIECTASIS		1 (10%)	2 (4%)	1 (2%)
#ADRENAL CORTEX	(5)	(10)	(49)	(48)
DEGENERATION, CYSTIC			7 (14%)	1 (2%)
METAMORPHOSIS FATTY				2 (4%)
HYPERPLASIA, FOCAL		1 (10%)	1 (2%)	
ANGIECTASIS				4 (8%)
#THYROID	(5)	(10)	(48)	(49)
CYSTIC FOLLICLES			3 (6%)	
HYPERPLASIA, C-CELL			5 (10%)	3 (6%)
HYPERPLASIA, FOLLICULAR-CELL			3 (6%)	
#THYROID FOLLICLE	(5)	(10)	(48)	(49)
ATROPHY, NOS			1 (2%)	
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(5)	(10)	(50)	(50)
HYPERPLASIA, NOS		1 (10%)	4 (8%)	
DYSPLASIA, NOS				1 (2%)
FIBROSING ADENOSIS		1 (10%)		
*MAMMARY LOBULE	(5)	(10)	(50)	(50)
HYPERPLASIA, NOS		1 (10%)		
#UTERUS	(5)	(10)	(48)	(46)
HYDROMETRA				1 (2%)
INFLAMMATION, CHRONIC				1 (2%)
#OVARY	(5)	(10)	(49)	(48)
FOLLICULAR CYST, NOS			1 (2%)	
NERVOUS SYSTEM				
NONE				
SPECIAL SENSE ORGANS				
NONE				
MUSCULOSKELETAL SYSTEM				
NONE				

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

TABLE C2. FEMALE RATS: NONNEOPLASTIC LESIONS (CONTINUED)

	HIGH DOSE CONTROL	LOW DOSE CONTROL	LOW DOSE	HIGH DOSE
BODY CAVITIES				
*MESENTERY	(5)	(10)	(50)	(50)
PERIARTERITIS				1 (2%)
ALL OTHER SYSTEMS				
NONE				
SPECIAL MORPHOLOGY SUMMARY				
NO LESION REPORTED	2	2	8	6

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

APPENDIX D

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MICE FED MALATHION IN THE DIET

TABLE D1.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE FED MALATHION IN THE DIET

	CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	10	50	50
ANIMALS NECROPSIED	10	48	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY	10	48	49
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG	(9)	(48)	(49)
INFLAMMATION, NOS		1 (2%)	
HEMATOPOIETIC SYSTEM			
#LYMPH NODE	(8)	(43)	(35)
INFLAMMATION, FOCAL		1 (2%)	
#MESENTERIC L. NODE	(8)	(43)	(35)
INFLAMMATION, CHRONIC		1 (2%)	
HYPERPLASIA, LYMPHOID		1 (2%)	2 (6%)
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#LIVER	(10)	(48)	(49)
INFLAMMATION, CHRONIC		1 (2%)	
DEGENERATION PARENCHYMATOUS			1 (2%)
NECROSIS, NOS			2 (4%)
#PEYERS PATCH	(10)	(44)	(46)
INFLAMMATION, NOS		1 (2%)	
HYPERPLASIA, LYMPHOID		1 (2%)	1 (2%)

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

TABLE D1. MALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
URINARY SYSTEM			
#KIDNEY	(10)	(47)	(49)
INFLAMMATION, CHRONIC FOCAL			1 (2%)
#KIDNEY/CORTEX	(10)	(47)	(49)
INFARCT, NOS		1 (2%)	
ENDOCRINE SYSTEM			
#PITUITARY	(6)	(42)	(40)
CYST, NOS		1 (2%)	
REPRODUCTIVE SYSTEM			
#TESTIS	(10)	(48)	(46)
ATROPHY, NOS	1 (10%)		
NERVOUS SYSTEM			
NONE			
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 D1. MALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	6	33	25
AUTOLYSIS/NO NECROPSY		2	1

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

TABLE D2.

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE FED MALATHION IN THE DIET

	CONTROL	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	10	50	50
ANIMALS NECROPSIED	10	49	48
ANIMALS EXAMINED HISTOPATHOLOGICALLY	10	49	47
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LUNG	(9)	(49)	(47)
INFLAMMATION, NOS			1 (2%)
HYPERPLASIA, ALVEOLAR EPITHELIUM		1 (2%)	
HEMATOPOIETIC SYSTEM			
#SPLEEN	(10)	(48)	(46)
INFLAMMATION, NOS			1 (2%)
HYPERPLASIA, LYMPHOID	1 (10%)	1 (2%)	
#HEPATIC LYMPH NODE	(10)	(43)	(38)
INFLAMMATION, GRANULOMATOUS	1 (10%)		
#THYMUS		(5)	(1)
HYPERPLASIA, LYMPHOID			1 (100%)
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#LIVER	(10)	(49)	(47)
DEGENERATION PARENCHYMATOUS			1 (2%)
*BILE DUCT	(10)	(49)	(48)
INFLAMMATION, CHRONIC			1 (2%)

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

TABLE D2. FEMALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
#PEYERS PATCH	(10)	(47)	(45)
HYPERPLASIA, LYMPHOID			1 (2%)
URINARY SYSTEM			
NONE			
ENDOCRINE SYSTEM			
NONE			
REPRODUCTIVE SYSTEM			
#UTERUS	(9)	(47)	(42)
HYDROMETRA			3 (7%)
PYOMETRA		1 (2%)	
INFLAMMATION, CHRONIC			1 (2%)
#UTERUS/ENDOMETRIUM	(9)	(47)	(42)
INFLAMMATION, SUPPURATIVE		1 (2%)	
DEGENERATION, CYSTIC			1 (2%)
HYPERPLASIA, CYSTIC	1 (11%)	12 (26%)	10 (24%)
#OVARY	(10)	(48)	(45)
FOLLICULAR CYST, NOS		3 (6%)	2 (4%)
INFLAMMATION, NOS	4 (40%)		
INFLAMMATION, SUPPURATIVE		2 (4%)	
NECROSIS, NOS	1 (10%)		
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
NONE			
MUSCULOSKELETAL SYSTEM			
NONE			

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

TABLE D2. FEMALE MICE: NONNEOPLASTIC LESIONS (CONTINUED)

	CONTROL	LOW DOSE	HIGH DOSE
BODY CAVITIES			
NONE			
ALL OTHER SYSTEMS			
NONE			
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	4	29	25
AUTO/NECROPSY/NO HISTO			1
AUTOLYSIS/NO NECROPSY		1	2

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

APPENDIX E

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS IN RATS FED MALATHION IN THE DIET

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Fed Malathion in the Diet[a]

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Pituitary: Adenoma, NOS (not otherwise specified) or Carcinoma[b]	0/10(0.00)	0/46(0.00)	3/43(0.07)	1/46(0.02)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.155	0.013
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			Infinite	Infinite
Lower Limit			0.646	0.054
Upper Limit			Infinite	Infinite
Weeks to First Observed Tumor	--	--	113	109
Pituitary: Chromophobe Adenoma[b]	0/10(0.00)	6/46(0.13)	6/43(0.14)	3/46(0.07)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.415	0.145
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			1.070	0.500
Lower Limit			0.308	0.086
Upper Limit			3.693	2.186
Weeks to First Observed Tumor	--	--	99	102

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Thyroid: Follicular-cell Adenoma or Carcinoma[b]	1/14(0.07)	4/46(0.09)	3/41(0.07)	7/47(0.15)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			1.024	2.085
Lower Limit			0.094	0.314
Upper Limit			52.448	91.743
Relative Risk (Pooled Control)[f]			0.841	1.713
Lower Limit			0.131	0.469
Upper Limit			4.671	7.483
Weeks to First Observed Tumor	108	--	113	100
Thyroid: C-cell Adenoma[b]	0/14(0.00)	2/46(0.04)	1/41(0.02)	3/47(0.06)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.019	0.192
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			0.561	1.468
Lower Limit			0.010	0.176
Upper Limit			10.353	16.917
Weeks to First Observed Tumor	--	--	113	109

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Pancreatic Islets: Islet-cell Adenoma[b]	0/15(0.00)	2/53(0.04)	1/45(0.02)	3/49(0.06)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.019	0.195
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			0.589	1.622
Lower Limit			0.010	0.194
Upper Limit			10.912	18.739
Weeks to First Observed Tumor	--	--	113	100
Adrenal Gland: Cortical Adenoma[b]	0/15(0.00)	2/52(0.04)	2/46(0.04)	0/50(0.00)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	--
Lower Limit			0.102	--
Upper Limit			Infinite	--
Relative Risk (Pooled Control)[f]			1.130	0.000
Lower Limit			0.086	0.000
Upper Limit			15.029	3.516
Weeks to First Observed Tumor	--	--	113	--

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Adrenal: Pheochromocytoma[b]	0/15(0.00)	2/52(0.04)	0/46(0.00)	3/50(0.06)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			--	Infinite
Lower Limit			--	0.192
Upper Limit			--	Infinite
Relative Risk (Pooled Control)[f]			0.000	1.560
Lower Limit			0.000	0.186
Upper Limit			3.814	18.027
Weeks to First Observed Tumor	--	--	113	100
Kidney: Mixed Tumor, Malignant[b]	0/15(0.00)	0/53(0.00)	2/48(0.04)	1/50(0.02)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.098	0.016
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			Infinite	Infinite
Lower Limit			0.327	0.056
Upper Limit			Infinite	Infinite
Weeks to First Observed Tumor	--	--	100	100

Table E1. Analyses of the Incidence of Primary Tumors in Male Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Liver: Neoplastic Nodule or Hepatocellular Carcinoma[b]	0/14(0.00)	1/53(0.02)	2/47(0.04)	1/50(0.02)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.093	0.016
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			2.255	1.060
Lower Limit			0.121	0.014
Upper Limit			130.193	81.565
Weeks to First Observed Tumor	--	--	113	109

[a]Dosed groups received time-weighted average doses of 4,700 or 8,150 ppm in feed.

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

[c]Beneath the incidence of tumors in a control group is the probability level for the Cochran-Armitage test when $P < 0.05$; otherwise, not significant (N.S.) is indicated. 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 matched-control group (*) or with the pooled-control group (**) when $P < 0.05$ for either control group; 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 probability level for departure from linear trend is given when $P < 0.05$ for any comparison.

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

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Fed Malathion in the Diet[a]

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Pituitary: Adenoma, NOS (not otherwise specified) or Carcinoma[b]	0/15(0.00)	1/46(0.02)	2/42(0.05)	1/45(0.02)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.112	0.019
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			2.190	1.022
Lower Limit			0.120	0.013
Upper Limit			126.082	78.492
Weeks to First Observed Tumor	--	--	105	109
Pituitary: Chromophobe Adenoma[b]	3/15(0.20)	9/46(0.20)	8/42(0.19)	7/45(0.16)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			0.952	0.778
Lower Limit			0.277	0.214
Upper Limit			5.086	4.276
Relative Risk (Pooled Control)[f]			0.974	0.795
Lower Limit			0.358	0.275
Upper Limit			2.571	2.187
Weeks to First Observed Tumor	98	--	108	102

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Thyroid: Follicular-cell Adenoma or Carcinoma[b]	0/15(0.00)	0/46(0.00)	0/48(0.00)	4/49(0.08)
P Values[c,d]	N.S.	P = 0.026	N.S.	N.S.
Relative Risk (Matched Control)[f]			--	Infinite
Lower Limit			--	0.301
Upper Limit			--	Infinite
Relative Risk (Pooled Control)[f]			--	Infinite
Lower Limit			--	0.872
Upper Limit			--	Infinite
Weeks to First Observed Tumor	--	--	--	105
Thyroid: C-cell Adenoma[b]	0/15(0.00)	1/46(0.02)	1/48(0.02)	2/49(0.04)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.018	0.096
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			0.958	1.878
Lower Limit			0.012	0.101
Upper Limit			73.689	108.485
Weeks to First Observed Tumor	--	--	113	109

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Pancreatic Islet: Islet-cell Adenoma[b]	0/15(0.00)	0/53(0.00)	2/50(0.04)	0/50(0.00)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Departure from Linear Trend[e]		P = 0.042		
Relative Risk (Matched Control)[f]			Infinite	--
Lower Limit			0.094	--
Upper Limit			Infinite	--
Relative Risk (Pooled Control)[f]			Infinite	--
Lower Limit			0.315	--
Upper Limit			Infinite	--
Weeks to First Observed Tumor	--	--	113	--
Adrenal Gland: Cortical Adenoma[b]	0/15(0.00)	0/50(0.00)	3/49(0.06)	0/48(0.00)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Departure from Linear Trend[e]		P = 0.014		
Relative Risk (Matched Control)[f]			Infinite	--
Lower Limit			0.195	--
Upper Limit			Infinite	--
Relative Risk (Pooled Control)[f]			Infinite	--
Lower Limit			0.615	--
Upper Limit			Infinite	--
Weeks to First Observed Tumor	--	--	113	--

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Liver: Neoplastic Nodule or Hepatocellular Carcinoma[b]	0/15(0.00)	1/53(0.02)	0/50(0.00)	3/49(0.06)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			--	Infinite
Lower Limit			--	0.196
Upper Limit			--	Infinite
Relative Risk (Pooled Control)[f]			0.000	3.245
Lower Limit			0.000	0.271
Upper Limit			19.777	166.782
Weeks to First Observed Tumor	--	--	--	109
Mammary Gland: Fibroadenoma[b]	0/15(0.00)	8/54(0.15)	6/50(0.12)	7/50(0.14)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			Infinite	Infinite
Lower Limit			0.513	0.621
Upper Limit			Infinite	Infinite
Relative Risk (Pooled Control)[f]			0.810	0.945
Lower Limit			0.249	0.313
Upper Limit			2.468	2.758
Weeks to First Observed Tumor	--	--	61	80

Table E2. Analyses of the Incidence of Primary Tumors in Female Rats Fed Malathion in the Diet[a]

(continued)

[a]Dosed groups received time-weighted average doses of 4,700 or 8,150 ppm in feed.

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

[c]Beneath the incidence of tumors in a control group is the probability level for the Cochran-Armitage test when $P < 0.05$; otherwise, not significant (N.S.) is indicated. 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 matched-control group (*) or with the pooled-control group (**) when $P < 0.05$ for either control group; 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 probability level for departure from linear trend is given when $P < 0.05$ for any comparison.

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

APPENDIX F

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS IN MICE FED MALATHION IN THE DIET

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Fed Malathion in the Diet[a]

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Liver: Neoplastic Nodule[b]	0/10(0.00)	3/49(0.06)	0/48(0.00)	6/49(0.12)
P Values[c,d]	P = 0.016	N.S.	N.S.	N.S.
Departure from Linear Trend[e]		P = 0.030		
Relative Risk (Matched Control)[f]			--	Infinite
Lower Limit			--	0.364
Upper Limit			--	Infinite
Relative Risk (Pooled Control)[f]			0.000	2.000
Lower Limit			0.000	0.455
Upper Limit			1.695	11.748
Weeks to First Observed Tumor	--	--	--	95
Liver: Hepatocellular Carcinoma[b]	2/10(0.20)	5/49(0.10)	7/48(0.15)	11/49(0.22)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			0.729	1.122
Lower Limit			0.180	0.320
Upper Limit			6.704	9.678
Relative Risk (Pooled Control)[f]			1.429	2.200
Lower Limit			0.419	0.767
Upper Limit			5.331	7.496
Weeks to First Observed Tumor	94	--	94	58

Table F1. Analyses of the Incidence of Primary Tumors in Male Mice Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Liver: Neoplastic Nodule or Hepatocellular Carcinoma[b]	2/10(0.20)	8/49(0.16)	7/48(0.15)	17/49(0.35)
P Values[c,d]	P = 0.041	P = 0.019	N.S.	P = 0.031**
Relative Risk (Matched Control)[f]			0.729	1.735
Lower Limit			0.180	0.540
Upper Limit			6.704	14.271
Relative Risk (Pooled Control)[f]			0.893	2.125
Lower Limit			0.300	0.967
Upper Limit			2.594	5.122
Weeks to First Observed Tumor	94	--	94	58

[a]Dosed groups received doses of 8,000 or 16,000 ppm in feed.

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

[c]Beneath the incidence of tumors in a control group is the probability level for the Cochran-Armitage test when P < 0.05; otherwise, not significant (N.S.) is indicated. 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 matched-control group (*) or with the pooled-control group (**) when P < 0.05 for either control group; 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 probability level for departure from linear trend is given when P < 0.05 for any comparison.

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

APPENDIX G

ANALYSIS OF FORMULATED DIETS FOR CONCENTRATIONS OF MALATHION

Table F2. Analyses of the Incidence of Primary Tumors in Female Mice Fed Malathion in the Diet[a]

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Liver: Neoplastic Nodule[b]	0/10(0.00)	1/48(0.02)	0/49(0.00)	2/47(0.04)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			--	Infinite
Lower Limit			--	0.069
Upper Limit			--	Infinite
Relative Risk (Pooled Control)[f]			0.000	2.043
Lower Limit			0.000	0.111
Upper Limit			18.270	117.919
Weeks to First Observed Tumor	--	--	--	95
Liver: Hepatocellular Carcinoma[b]	0/10(0.00)	2/48(0.04)	0/49(0.00)	0/47(0.00)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			--	--
Lower Limit			--	--
Upper Limit			--	--
Relative Risk (Pooled Control)[f]			0.000	0.000
Lower Limit			0.000	0.000
Upper Limit			3.309	3.447
Weeks to First Observed Tumor	--	--	--	--

Table F2. Analyses of the Incidence of Primary Tumors in Female Mice Fed Malathion in the Diet[a]

(continued)

Topography: Morphology	Matched Control	Pooled Control	Low Dose	High Dose
Liver: Neoplastic Nodule or Hepatocellular Carcinoma[b]	0/10(0.00)	3/48(0.06)	0/49(0.00)	2/47(0.04)
P Values[c,d]	N.S.	N.S.	N.S.	N.S.
Relative Risk (Matched Control)[f]			--	Infinite
Lower Limit			--	0.069
Upper Limit			--	Infinite
Relative Risk (Pooled Control)[f]			0.000	0.681
Lower Limit			0.000	0.059
Upper Limit			1.628	5.673
Weeks to First Observed Tumor	--	--	--	95

[a]Dosed groups received doses of 8,000 or 16,000 ppm in feed.

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

[c]Beneath the incidence of tumors in a control group is the probability level for the Cochran-Armitage test when $P < 0.05$; otherwise, not significant (N.S.) is indicated. 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 matched-control group (*) or with the pooled-control group (**) when $P < 0.05$ for either control group; 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 probability level for departure from linear trend is given when $P < 0.05$ for any comparison.

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

APPENDIX G

Analysis of Formulated Diets for Concentrations of Malathion

A 10-g sample of the diet mixture was shaken with 125 ml hexane at room temperature for 16 hours, then filtered through Celite with hexane washes, and reduced in volume to 10 ml. After appropriate dilutions, the solution was quantitatively analyzed for malathion by gas-liquid chromatography (electron-capture detector, 10% DC-200 on Gas-Chrom Q column). Recoveries were checked with spiked samples, and external standards were used for calibration.

Theoretical Concentrations in Diet (ppm)	No. of Samples	Sample Analytical Mean (ppm)	Coefficient of Variation (%)	Range (ppm)
4,000	20	4,098	4.9%	3,690-4,570
8,000	35	8,188	4.2%	7,390-8,880
16,000	25	16,290	5.0%	14,950-17,790

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

January 18, 1978

The Clearinghouse on Environmental Carcinogens was established in May, 1976 under the authority of the National Cancer Act of 1971 (P.L. 92-218). The purpose of the Clearinghouse is to advise on the National Cancer Institute's bioassay program to identify and 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 organic chemistry, biochemistry, biostatistics, toxicology, pathology, and epidemiology. Representatives of various environmental 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 NCI bioassay reports on chemicals studied for carcinogenicity. In this context, below is the edited excerpt from the minutes of the Subgroup's meeting at which Malathion was reviewed.

The primary reviewer agreed with the conclusion of the report that there was no clear evidence of an association between the tumor incidence in the treated animals and the administration of Malathion. He stressed the fact that the dose levels administered were sufficiently high to produce an effect. The primary reviewer was particularly critical of the small size of the matched control groups. He pointed out the incidence of hepatocellular carcinomas in the treated male mice, although it was not statistically significant.

The secondary reviewer questioned the validity of the bioassay in terms of its significance as a valid "negative" study. He pointed out the increased incidence of adenomas of the thyroid in the treated rats, as well as their earlier appearance in the animals at the high dose level. He argued this may indicate an important biological effect despite

the statistical insignificance of the finding. Considering this finding and the high human exposure, he suggested that the study be repeated using a different route of exposure.

A Program staff pathologist commented that the time to the detection of the thyroid tumors in the treated animals may be misleading. Since it was not possible to detect the cause of the animals' deaths, he noted that the difference in survival time between the treated and control animals may not be truly significant. It was the feeling of the Program staff that there was no evidence to indicate the carcinogenicity of Malathion, although there is always a probability of obtaining a false negative.

The Subgroup agreed unanimously to accept the report on the bioassay of Malathion.

Members present were:

Arnold Brown (Acting Chairman), Mayo Clinic
Lawrence Garfinkel, American Cancer Society
Joseph Highland, Environmental Defense Fund
Charles Kensler, Arthur D. Little Company
Verald K. Rowe, Dow Chemical, U.S.A.
Sheldon Samuels, Industrial Union Department, AFL-CIO
Louise Strong, University of Texas Health Sciences Center
Sidney Wolfe, Health Research Group

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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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