NATIONAL TOXICOLOGY PROGRAM
Technical Report Series
No. 281

TOXICOLOGY AND CARCINOGENESIS STUDIES OF HC RED NO. 3

[2,-((4-AMINO-2-NITROPHENYL)AMINO)ETHANOL]

(CAS NO. 2871-01-4)

IN F344/N RATS AND B6C3F$_1$ MICE

(GAVAGE STUDIES)

U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
Public Health Service
National Institutes of Health

NATIONAL TOXICOLOGY PROGRAM

The National Toxicology Program (NTP), established in 1978, develops and evaluates scientific information about potentially toxic and hazardous chemicals. This knowledge can be used for protecting the health of the American people and for the primary prevention of disease. By bringing together the relevant programs, staff, and resources from the U.S. Public Health Service, DHHS, the National Toxicology Program has centralized and strengthened activities relating to toxicology research, testing and test development/validation efforts, and the dissemination of toxicological information to the public and scientific communities and to the research and regulatory agencies.

The NTP is made up of four charter DHHS agencies: the National Cancer Institute (NCI), National Institutes of Health; the National Institute of Environmental Health Sciences (NIEHS), National Institutes of Health; the National Center for Toxicological Research (NCTR), Food and Drug Administration; and the National Institute for Occupational Safety and Health (NIOSH), Centers for Disease Control. In July 1981, the Carcinogenesis Bioassay Testing Program, NCI, was transferred to the NIEHS.

NTP TECHNICAL REPORT

ON THE

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NATIONAL TOXICOLOGY PROGRAM
P.O. Box 12233
Research Triangle Park, NC 27709

January 1986

NTP TR 281

NIH Publication No. 86-2537

U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
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NOTE TO THE READER

These studies are designed and conducted to characterize and evaluate the toxicologic potential, including carcinogenic activity, of selected chemicals in laboratory animals (usually two species, rats and mice). Chemicals selected for testing in the NTP Carcinogenesis Program are chosen primarily on the bases of human exposure, level of production, and chemical structure. Selection per se is not an indicator of a chemical's carcinogenic potential. Negative results, in which the test animals do not have a greater incidence of cancer than control animals, do not necessarily mean that a test chemical is not a carcinogen, inasmuch as the experiments are conducted under a limited set of conditions. Positive results demonstrate that a test chemical is carcinogenic for animals under the conditions of the test and indicate that exposure to the chemical has the potential for hazard to humans. The determination of the risk to humans from chemicals found to be carcinogenic in animals requires a wider analysis which extends beyond the purview of this study.

Five categories of interpretative conclusions were adopted for use in June 1983 in the Technical Reports series to specifically emphasize consistency and the concept of actual evidence of carcinogenicity. For each definitive study result (male rats, female rats, male mice, female mice), one of the following quintet will be selected to describe the findings. These categories refer to the strength of the experimental evidence and not to either potency or mechanism.

- **Clear Evidence of Carcinogenicity** is demonstrated by studies that are interpreted as showing a chemically related increased incidence of malignant neoplasms, studies that exhibit a substantially increased incidence of benign neoplasms, or studies that exhibit an increased incidence of a combination of malignant and benign neoplasms where each increases with dose.
- **Some Evidence of Carcinogenicity** is demonstrated by studies that are interpreted as showing a chemically related increased incidence of benign neoplasms, studies that exhibit marginal increases in neoplasms of several organs/tissues, or studies that exhibit a slight increase in uncommon malignant or benign neoplasms.
- **Equivocal Evidence of Carcinogenicity** is demonstrated by studies that are interpreted as showing a chemically related marginal increase of neoplasms.
- **No Evidence of Carcinogenicity** is demonstrated by studies that are interpreted as showing no chemically related increases in malignant or benign neoplasms.
- **Inadequate Study of Carcinogenicity** demonstrates that because of major qualitative or quantitative limitations, the studies cannot be interpreted as valid for showing either the presence or absence of a carcinogenic effect.

Additionally, the following concepts (as patterned from the International Agency for Research on Cancer Monographs) have been adopted by the NTP to give further clarification of these issues:

> The term *chemical carcinogenesis* generally means the induction by chemicals of neoplasms not usually observed, the earlier induction by chemicals of neoplasms that are commonly observed, or the induction by chemicals of more neoplasms than are generally found. Different mechanisms may be involved in these situations. Etymologically, the term *carcinogenesis* means induction of cancer, that is, of malignant neoplasms; however, the commonly accepted meaning is the induction of various types of neoplasms or of a combination of malignant and benign neoplasms. In the Technical Reports, the words *tumor* and *neoplasm* are used interchangeably.

This study was initiated by the National Cancer Institute's Carcinogenesis Bioassay Program, now part of the National Institute of Environmental Health Sciences, National Toxicology Program. The studies described in this Technical Report have been conducted in compliance with NTP chemical health and safety requirements and must meet or exceed all applicable Federal, state, and local health and safety regulations. All NTP toxicology and carcinogenesis studies are subjected to a data audit before being presented for peer review.

Although every effort is made to prepare the Technical Reports as accurately as possible, mistakes may occur. Readers are requested to identify any mistakes so that corrective action may be taken. Further, anyone who is aware of related ongoing or published studies not mentioned in this report is encouraged to make this information known to the NTP. Comments and questions about the National Toxicology Program Technical Reports on Toxicology and Carcinogenesis Studies should be directed to Dr. J.E. Huff, National Toxicology Program, P.O. Box 12233, Research Triangle Park, NC 27709 (919-541-3780).

These NTP Technical Reports are available for sale from the National Technical Information Service, U.S. Department of Commerce, 5285 Port Royal Road, Springfield, VA 22161 (703-487-4650). Single copies of this Technical Report are available without charge (and while supplies last) from the NTP Public Information Office, National Toxicology Program, P.O. Box 12233, Research Triangle Park, NC 27709.

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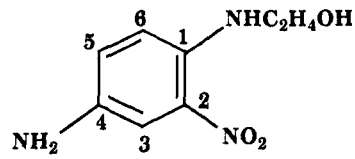

HC RED NO. 3

2-((4-AMINO-2-NITROPHENYL)AMINO)ETHANOL

$C_8H_{11}N_3O_3$ Molecular weight 197.2

ABSTRACT

Toxicology and carcinogenesis studies of HC Red No. 3 (97% pure), a semipermanent hair dye, were conducted by administering the chemical in corn oil by gavage for 105 weeks to groups of 50 male and 50 female F344/N rats and for 104 weeks to groups of 50 male and 50 female B6C3F$_1$ mice. The dosage regimen used for rats was 0, 250, or 500 mg/kg per day and for mice, 0, 125, or 250 mg/kg per day. Doses were administered 5 days per week. In prior 13-week studies, these doses produced no signs of toxicity when administered 5 days per week.

In the 2-year studies, the administration of HC Red No. 3 did not affect body weight gains of male or female rats or mice. Body weight gains by all groups of female mice were reduced because of a reproductive tract infection. Survival of male and female rats and mice was not reduced by administration of HC Red No. 3. The survival of female mice, including vehicle controls, was reduced relative to historical survival rates due to a reproductive tract infection. The infection, accompanied by weight loss, high mortality, and suppurative inflammation of multiple organs, was found in 36/50 vehicle control, 32/50 low dose, and 29/50 high dose female mice. *Klebsiella pneumoniae* was isolated from infected tissues.

Pigmentation of various tissues in both rats and mice was a common observation in both the 13-week and the 2-year studies. The pigment was not identified but was presumed to be a derivative of HC Red No. 3. Very minimal nephropathy was found in dosed female rats, but its relationship to HC Red No. 3 is equivocal. Mild nephrosis was found in dosed female mice, but this effect may have been secondary to the infection of the genital tract.

There was an increase in the incidence of mammary gland fibroadenomas or cystadenomas in low dose female rats. The incidence of this lesion in high dose female rats was not increased (vehicle control, 14/50, 28%; low dose, 25/50, 50%; high dose, 11/50, 22%). Largely because of the lack of a dose response, the increased incidence in the low dose females was not considered to be due to HC Red No. 3. No increased incidences of neoplasms were seen in male rats.

Transitional cell papillomas of the urinary bladder were detected in one high dose male rat, two low dose female rats, and one high dose female rat; none was observed in the vehicle controls. These uncommon neoplasms were found in animals that survived to the termination of the study and were not accompanied by other proliferative lesions.

The incidence of hepatocellular adenomas or carcinomas (combined) was increased in high dose male mice, whereas the incidence of these neoplasms in low dose male mice was significantly lower than that in the vehicle controls (25/50; 15/50; 35/50). Hepatocellular carcinomas in three vehicle control, one low dose, and five high dose male mice metastasized to the lung. The incidences of liver neoplasms in dosed female mice were not significantly different from those in the vehicle control group.

HC Red No. 3 was mutagenic in *Salmonella typhimurium* strains TA97, TA98, and TA100, but not in TA1535, in the presence or absence of Aroclor 1254-induced male Sprague-Dawley rat or male Syrian hamster liver S9 when tested by the preincubation protocol.

An audit of the experimental data was conducted for these 2-year toxicology and carcinogenesis studies on HC Red No. 3. No data discrepancies were found that influenced the final interpretations.

Under the conditions of these 2-year gavage studies of HC Red No.3, there was *no evidence of carcinogenicity** for male or female F344/N rats given 250 or 500 mg/kg per day. There was *equivocal evidence of carcinogenicity* for male $B6C3F_1$ mice as indicated by an increased incidence of hepatocellular adenomas or carcinomas (combined) in the 250 mg/kg group. Poor survival coupled with lack of significant findings rendered the study in female $B6C3F_1$ mice an *inadequate study of carcinogenicity*. Both sexes of both species may have been able to tolerate higher doses of HC Red No. 3. Therefore, the sensitivity of these studies for detecting carcinogenesis may have been limited.

*Categories of evidence of carcinogenicity are defined in the Note to the Reader on page 2.

CONTRIBUTORS

The NTP Technical Report on the Toxicology and Carcinogenesis Studies of HC Red No. 3 is based on the 13-week studies that began in February 1979 and ended in May 1979 and on the 2-year studies that began in November 1979 and ended in November 1981 at Southern Research Institute.

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The members of the Peer Review Panel who evaluated the draft Technical Report on HC Red No. 3 on March 29, 1985, are listed below. Panel members serve as independent scientists, not as representatives of any institution, company, or governmental agency. In this capacity, Panel members have five major responsibilities: (a) to ascertain that all relevant literature data have been adequately cited and interpreted, (b) to determine if the design and conditions of the NTP studies were appropriate, (c) to ensure that the Technical Report presents the experimental results and conclusions fully and clearly, (d) to judge the significance of the experimental results by scientific criteria, and (e) to assess the evaluation of the evidence of carcinogenicity and other observed toxic responses.

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SUMMARY OF PEER REVIEW COMMENTS ON THE TOXICOLOGY AND CARCINOGENESIS STUDIES OF HC RED NO. 3

On March 29, 1985, the draft Technical Report on the toxicology and carcinogenesis studies of HC Red No. 3 received peer review by the National Toxicology Program Board of Scientific Counselors' Technical Reports Review Subcommittee and associated Panel of Experts. The review meeting began at 9:00 a.m. in the Conference Center, Building 101, National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina.

Dr. Kotelchuck, a principal reviewer, agreed in principle with the conclusions. He stated that the results of the short-term studies indicated that the high dose used in the 2-year studies was well below the dose that could have been easily tolerated in both sexes of both species. Thus, the conclusions should reflect this.

As a second principal reviewer, Dr. Tannenbaum said he agreed with the use of the gavage route over dermal exposure but asked that the discussion indicate that metabolism by the two routes could be quite different; for example, nitrophenylenediamine dyes are metabolized in the gastrointestinal tract. He was pleased to note that nitrosamines were analyzed but said more information was needed on methods of analysis, levels found, and possible biologic effects of these contaminants. Dr. Mennear, NTP, said that the discussion would be expanded on route-specific metabolism and on the nitrosamines [see page 54].

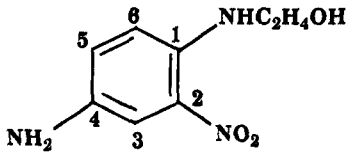
As a third principal reviewer, Dr. Hooper disagreed with the evidence categories in male and female rats and male mice because these animals could have tolerated higher doses as shown by no effects on body weight or survival. Further, in recently completed NTP studies of the structurally related dyes HC Blue No. 1 and HC Blue No. 2, much higher doses in mice were well tolerated. Dr. Hooper proposed that there be two categories: one referring to the strength of evidence, the second referring to the adequacy of the study design. For example, the conclusions in male mice could be that this was an inadequate study of carcinogenicity producing equivocal evidence of a carcinogenic effect. At the least, he suggested that the study design was inadequate because low doses were used for all four sex/species groups. Dr. McConnell, NIEHS/NTP, replied that increasing the number of categories or adding qualifiers to the existing ones is not necessary, and Dr. Huff, NIEHS/NTP, noted there were already qualifiers for the stated conclusions on male rats and mice that higher doses may have been tolerated. Dr. Hooper proposed that the conclusions as written be accepted with a conditional modifying statement noting that the sensitivity of this study for detecting a carcinogenic effect may have been limited by poor survival (female mice) or by administration of less than a maximum tolerated dose (rats and male mice). Dr. Turnbull seconded the motion.

In subsequent discussion, Dr. Kociba and Dr. Purchase questioned the interpretation of equivocal evidence of carcinogenicity based on the data for mammary gland tumors in female rats. Despite the high incidence in the low dose group, this tumor was common and variable, and the incidence in the high dose group was lower than that in the concurrent control group; thus, there was no biologic basis for even a marginal effect of administration. Dr. Haseman, NIEHS, noted that the low-dose rate, however, was well above the historical range for gavage controls.

Dr. Hook said he would accept a motion for an amendment to the previous motion having to do with the conclusion in rats. Dr. Kociba moved that the original motion be amended so that the conclusion read: "...there was *no evidence of carcinogenicity* for male and female rats." Dr. Purchase seconded the motion, and it was approved by six affirmative votes; there were four negative votes (Dr. Harper, Dr. Hooper, Dr. Kotelchuck, and Dr. Perera). Dr. Hook asked for a vote on Dr. Hooper's motion to include Dr. Kociba's amendment with a modifying statement indicating that higher doses might have been tolerated in all four experiments. The motion was approved unanimously.

I. INTRODUCTION

I. INTRODUCTION



HC RED NO. 3

2-((4-AMINO-2-NITROPHENYL)AMINO)ETHANOL

$C_8H_{11}N_3O_3$ Molecular weight 197.2

HC Red No. 3 is a nitrophenylenediamine derivative used exclusively as a semipermanent hair dye. Approximately 5,000-10,000 pounds of HC Red No. 3 are used annually.

Semipermanent hair color products are generally shampoo-in preparations that are applied to the hair, lathered, and then allowed to remain in contact with the hair (and scalp) for 30-45 minutes (Frenkel and Brody, 1973). The concentration of HC Red No. 3 used in these preparations ranges from 0.1% to 5%.

Past studies in which HC Red No. 3 was administered to laboratory animals were conducted on complex mixtures of dyes, dye intermediates, and product base chemicals (solvents and detergents). Wernick et al. (1975) administered a composite of 15 semipermanent hair dyes, formulated in product base materials, to dogs, rats, and rabbits. The composite contained 6.95% dye chemicals, including 0.02% HC Red No. 3. The mixture was tested for systemic effects in beagle dogs (dietary administration for 2 years), for teratologic effects in Sprague-Dawley rats (dietary administration on days 6 through 15 of gestation) and New Zealand rabbits (gavage administration on days 6 through 18 of gestation), and for reproductive effects in Sprague-Dawley rats (dietary administration). The largest doses of HC Red No. 3 delivered by the mixture were 0.02 mg/kg per day to dogs and rabbits and 0.16 mg/kg per day (estimated) to rats. No compound-related effects were observed.

No studies on the absorption, distribution, metabolism, excretion, or genetic toxicology of HC Red No. 3 have been published. However, the NTP found that HC Red No. 3 was mutagenic in *Salmonella typhimurium* strains TA97, TA98, and TA100, but not in TA1535, in the presence or absence of Aroclor 1254-induced male Sprague-Dawley rat or Syrian hamster liver S9 when tested according to the preincubation protocol (Appendix M). The results show that, in Salmonella, HC Red No. 3 is a mutagen whose mutagenic activity is greatly enhanced by liver S9 metabolism.

The International Agency for Research on Cancer published a monograph on aromatic amines, including hair dye preparations (IARC, 1982). The epidemiologic information concerning relationships between various human cancers and either employment as a hairdresser or the personal use of hair dyes was evaluated as inconclusive.

HC Red No. 3 is one of five semipermanent hair dyes selected for toxicologic and carcinogenicity assessment in a chemical class study of hair color materials. The other dyes studied were HC Blue No. 1, HC Blue No. 2, C.I. Disperse Blue No. 1, and C.I. Acid Orange No. 3.

The results of the studies of HC Blue No. 1 (NTP, 1985a) and HC Blue No. 2 (NTP, 1985b) have been reported, and the other studies are currently in progress. The structures for HC Red No. 3, HC Blue No. 1, and HC Blue No. 2 are compared in the Discussion and Conclusions

(Chapter IV). HC Blue No. 1 caused hepatocellular carcinomas in mice, liver neoplasms in male rats, alveolar/bronchiolar neoplasms in female rats, and thyroid gland neoplasms in male mice. HC Blue No. 2 did not cause any increased incidences of neoplasms in either rats or mice.

During the planning of this series of studies on hair dyes, it was felt that, regardless of the degree of dermal absorption of dye, a larger proportion of chemical would be absorbed through the gastrointestinal tract than through the skin. Therefore, the oral route of administration was selected for each chemical to provide a more rigorous test than would be possible through dermal application.

II. MATERIALS AND METHODS

PROCUREMENT AND CHARACTERIZATION OF HC RED NO. 3

PREPARATION AND CHARACTERIZATION OF DOSE MIXTURES

SINGLE-ADMINISTRATION STUDIES

FOURTEEN-DAY STUDIES

THIRTEEN-WEEK STUDIES

TWO-YEAR STUDIES

Study Design

Source and Specifications of Animals

Animal Maintenance

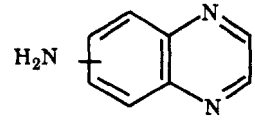
Clinical Examinations and Pathology

Statistical Methods

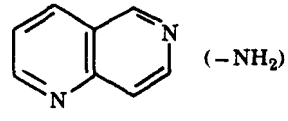
PROCUREMENT AND CHARACTERIZATION OF HC RED NO. 3

HC Red No. 3 [2-((4-amino-2-nitrophenyl)-amino)ethanol] was obtained from Clairol Research Laboratories (Stamford, Connecticut) in two lots (Table 1) as the unformulated technical-grade dye. Clairol reported that the dye was 97.3% pure. Purity and identity analyses on both lots were conducted at Midwest Research Institute (Kansas City, Missouri).

The identity of the material was confirmed by infrared, ultraviolet/visible, and nuclear magnetic resonance spectroscopy (Appendix G). All spectroscopic data were consistent with the structure of HC Red No. 3. Purity of the two batches was determined by elemental analysis, water analysis, titration of one amine group, thin-layer chromatography, and high-performance liquid chromatography. Results of these analyses indicated that both batches of test material were greater than 97% pure; these findings were consistent with the manufacturer's specifications. High-performance liquid chromatographic data indicated that there were two impurities in each batch with areas greater than 1% relative to the major peak area. These impurities were isolated by high-performance liquid chromatography and identified by mass spectrometry. One impurity was identified as a heterocyclic fused-ring compound, probably an aminoquinoxaline (I) or an aminonaphthyridine (II).



I. AMINOQUINOXALINE

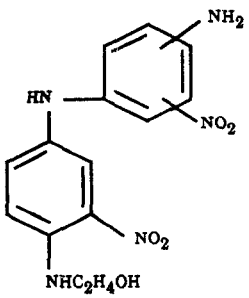


II. AMINONAPHTHYRIDINE

TABLE 1. IDENTITY AND SOURCE OF LOTS USED IN THE GAVAGE STUDIES OF HC RED NO. 3

Single-Administration Studies	Fourteen-Day Studies	Thirteen-Week Studies	Two-Year Studies
Lot Numbers			
5890377	5890377	5890377	5890377; C080480
Date of Initial Use of Each Lot			
4/11/78	N/A	N/A	11/27/79; 10/24/80
Supplier			
Clairol Research Labs (Stamford, CT)	Same as single-administration studies	Same as single-administration studies	Same as single-administration studies

The concentration of this impurity was approximately 1.1% for lot no. 5890377 and approximately 0.3% for lot no. C080480. The second impurity was identified as an analog of HC Red No. 3 (III) and was present at a concentration of approximately 0.8% for lot no. 5890377 and approximately 1.7% for lot no. C080480.



III. HC RED NO. 3 ANALOG

Samples of both lots of HC Red No. 3 were analyzed by Thermo Electron Corporation for possible nitrosamine impurities. High-performance liquid chromatography-thermal energy analysis of the test samples indicated that lot no. 5890377 contained approximately 20 ± 5 ppm total nitrosamines and lot no. C080480 contained approximately 11 ± 8 ppm total nitrosamines. The identities of the nitrosamines observed in these two test samples were not determined. The peaks attributed to the polar nitrosamines in each sample exhibited excessive tailing and may have been due to interferences from the nitro group of HC Red No. 3.

The testing laboratory stored the test material at 22° C. Results of periodic reanalysis of the chemical at Southern Research Institute by infrared and ultraviolet visible spectroscopy indicated that no detectable deterioration occurred over the course of the studies.

PREPARATION AND CHARACTERIZATION OF DOSE MIXTURES

In the single-administration and 14-day studies, appropriate amounts of HC Red No. 3 were mixed with 1% carboxymethyl-cellulose (CMC). The CMC was dissolved in saline in the single-administration studies and in water in the 14-day studies. In the 13-week and 2-year studies, HC Red No. 3 was mixed with corn oil (Table 2). In all studies, the mixtures were continually stirred with a magnetic stirrer while doses were being administered.

TABLE 2. PREPARATION AND STORAGE OF DOSE MIXTURES IN THE GAVAGE STUDIES OF HC RED NO. 3

Single-Administration Studies	Fourteen-Day Studies	Thirteen-Week Studies	Two-Year Studies
Preparation			
Appropriate amounts of carboxymethylcellulose (CMC) in saline and HC Red No. 3 were stirred in a beaker with a spatula; the mixture was then sonicated for 10 min (rats) or 5 min (mice) until a dark red suspension was obtained	Same as the single-administration studies except CMC was in water and HC Red No. 3/CMC mixture was sonicated for only 5 min	Appropriate amounts of HC Red No. 3 and corn oil were manually shaken for 1 min, then mixed with a magnetic stirrer until the suspension was uniform	Appropriate amounts of HC Red No. 3 and corn oil were mixed with a magnetic stirrer
Maximum Storage Time			
Animals dosed immediately after chemical/vehicle preparation	3-4 d	1 wk	7 d from 11/27/79 to 9/12/80, then 14 d
Storage Conditions			
None	Room temp	22° C	Room temp

Studies of potential vehicles for administering HC Red No. 3 indicated that the recovery of HC Red No. 3 from feed decreased with time and increasing temperature (Appendix H). The amount of the test chemical recovered decreased as storage temperatures increased, with a 21% decrease being observed after 3 hours at 25° C. HC Red No. 3 feed blends were found to be unstable at storage temperatures of 5° C and above. The underlying reason for this loss was not pursued.

HC Red No. 3 was found to be stable in either 1% aqueous methyl cellulose or corn oil (Appendix I). Preliminary studies with corn oil suspensions of HC Red No. 3 established that homogeneous suspensions could be prepared and that the suspended HC Red No. 3 was stable in corn oil for 7 days at room temperature. The testing laboratory later expanded the stability study and confirmed a 14-day stability of the HC Red No. 3 corn oil suspensions.

The dose preparation method that was employed in these studies consisted of suspending the HC Red No. 3 in corn oil on a weight/volume basis with a magnetic stirrer. The dose mixtures were prepared weekly by the testing laboratory and stored at 22° C until used for dosing. Biweekly mixing was performed after September 12, 1980.

Dose mixtures of HC Red No. 3 in corn oil were analyzed periodically by the testing and referee laboratories to confirm chemical content. The analytical method included a methanolic extraction and a spectrophotometric quantitation step (Appendix J). Because 7/49 mixtures sampled were not within 10% of the target concentration, it is estimated that doses were formulated within specifications 86% of the time during the 2-year studies (Table 3; Appendix K, Table K2).

SINGLE-ADMINISTRATION STUDIES

Male and female F344/N rats and B6C3F$_1$ mice were obtained from Harlan Industries and held for 14 days before the test began.

Groups of five rats of each sex were administered a single dose of 62, 125, 250, 500, or 1,000 mg/kg HC Red No. 3 in 1% carboxymethyl cellulose in saline by gavage. Groups of five mice of each sex were administered a single dose of 31, 62, 125, 250, or 500 mg/kg HC Red No. 3 in 1% carboxymethyl cellulose by gavage. Details of animal maintenance are given in Table 4.

FOURTEEN-DAY STUDIES

Male and female F344/N rats and B6C3F$_1$ mice were obtained from Harlan Industries and held for 14 days before the studies began. Animals were approximately 7-8 weeks old when placed on study.

Groups of five rats of each sex were administered HC Red No. 3 (0, 62, 125, 250, 500, or 1,000 mg/kg) in 1% aqueous carboxymethyl cellulose for 14 consecutive days. Groups of five mice of each sex were administered 0, 31, 62, 125, 250, or 500 mg/kg for 14 consecutive days.

Animals were housed five per cage and received water and feed ad libitum. Details of animal maintenance are presented in Table 4.

THIRTEEN-WEEK STUDIES

Thirteen-week studies were conducted to evaluate the cumulative toxicity of HC Red No. 3 and to determine the doses to be used in the 2-year studies. Five- to 6-week-old male and female F344/N rats and B6C3F$_1$ mice were obtained from Charles River Breeding Laboratories,

TABLE 3. SUMMARY OF RESULTS OF ANALYSIS OF DOSE MIXTURES IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

	Determined Concentration for Target Concentration of			
	1.25% (w/v)	2.50% (w/v)	5.00% (w/v)	10.00% (w/v)
Mean (percent, w/v)	1.22	2.48	4.93	10.10
Standard deviation	0.109	0.174	0.332	0.935
Coefficient of variation (percent)	8.9	7.0	6.7	9.3
Range (percent, w/v)	0.97-1.39	2.22-2.82	4.27-5.50	9.33-12.7
Number of samples	12	12	13	12

TABLE 4. EXPERIMENTAL DESIGN AND MATERIALS AND METHODS IN THE GAVAGE STUDIES OF HC RED NO. 3

Single-Administration Studies	Fourteen-Day Studies	Thirteen-Week Studies	Two-Year Studies
EXPERIMENTAL DESIGN			
Size of Test Groups			
5 males and 5 females of each species	5 males and 5 females of each species	10 males and 10 females of each species	50 males and 50 females of each species
Doses			
Rats--62, 125, 250, 500, or 1,000 mg/kg HC Red No. 3 in 1% carboxymethyl cellulose (CMC) in saline by gavage; dose vol: 5 ml/kg; mice--31, 62, 125, 250, or 500 mg/kg; dose vol: 10 ml/kg, except 500 mg/kg--20 ml/kg. Rats--13-gauge needle; mice--23-gauge needle	Rats--0, 62, 125, 250, 500, or 1,000 mg/kg HC Red No. 3 in 1% aqueous CMC by gavage; mice--0, 31, 62, 125, 250, or 500 mg/kg; dose vol: same as single-administration studies Rats--13-gauge needle; mice--23-gauge needle	Rats--0, 62, 125, 250, 500, or 1,000 mg/kg HC Red No. 3 in corn oil by gavage; dose vol: 5 ml/kg; mice--0, 15, 31, 62, 125, or 250 mg/kg; dose vol: 10 ml/kg. Rats--13-gauge needle; mice--23-gauge needle	Rats--0, 250, or 500 mg/kg HC Red No. 3 in corn oil by gavage; mice--0, 125, or 250 mg/kg; dose vol: same as 13-wk studies. Rats--13-gauge needle; mice--18-gauge needle
Date of First Dose			
4/11/78	9/20/78	1/31/79	11/27/79
Date of Last Dose			
N/A	10/3/78	4/30/79	Rats--11/27/81; mice--11/20/81
Duration of Dosing			
One time only	14 d	13 wk	Rats--5 d/wk for 105 wk; mice--5 d/wk for 104 wk
Type and Frequency of Observation			
Observed 2 × d for 15 d; weighed on d 0 and d 15	Observed 2 × d for 15 d; weighed on d 1 and d 15	Observed 2 × d; weighed 1 × wk; clinical exam 1 × wk	Observed 2 × d; weighed on d 0, 1 × wk for 12 wk, then once every 4 wk; palpated 1 × wk
Necropsy and Histologic Examination			
Not performed	Necropsies performed on all animals; histopath exam was not performed	Necropsies and histopath exam performed on all vehicle control, 1,000 mg/kg rats, and 250 mg/kg mice; the following tissues were examined: skin, mandibular lymph node, mammary gland, salivary gland, thigh muscle, femur including marrow, thymus, trachea, lungs and bronchi, heart, thyroid gland, stomach, parathyroids, esophagus, small intestine, colon, mesenteric lymph node, liver, pancreas, spleen, kidneys, adrenal glands, urinary bladder, vesicular gland/prostate/testis or ovary/uterus, brain, pituitary gland; kidneys and thyroid gland of rats administered 250 or 500 mg/kg were also examined microscopically	Necropsies and histopath exam performed. The following tissues of all animals were examined: gross lesions, skin, mandibular lymph node, mammary gland, salivary gland, thigh muscle, sciatic nerve, femur including marrow, thymus, costochondral junction, larynx, lungs and bronchi, trachea, heart, thyroid gland, parathyroids, esophagus, stomach, duodenum, jejunum, eyes, tissue masses with regional lymph node, ileum, colon, cecum, rectum, liver, mesenteric lymph node, inguinal lymph node, pancreas, spleen, kidneys, adrenal glands, urinary bladder, testis/epididymis/seminal vesicles/prostate or ovaries/uterus/fallopian

TABLE 4. EXPERIMENTAL DESIGN AND MATERIALS AND METHODS IN THE GAVAGE STUDIES OF HC RED NO. 3 (Continued)

Single-Administration Studies	Fourteen-Day Studies	Thirteen-Week Studies	Two-Year Studies
Necropsy and Histologic Examination (Continued)			
			tube/vagina, nasal cavity, brain, preputial gland, pituitary gland, spinal cord, gallbladder (mice), and external and middle ear
ANIMALS AND ANIMAL MAINTENANCE			
Testing Laboratory			
Southern Research Institute	Southern Research Institute	Southern Research Institute	Southern Research Institute
Species			
F344/N rats; $B6C3F_1$ mice	F344/N rats; $B6C3F_1$ mice	F344/N rats; $B6C3F_1$ mice	F344/N rats; $B6C3F_1$ mice
Animal Source			
Harlan Industries (Indianapolis, IN)	Harlan Industries (Indianapolis, IN)	Charles River Breeding Laboratories	Charles River Breeding Laboratories (Portage, MI)
Time Held Before Start of Test			
14 d	14 d	21 d	20 d
Age When Placed on Study			
7-8 wk	7-8 wk	7-8 wk	Rats--7-8 wk; mice--8 wk
Age When Killed			
9-10 wk	9-10 wk	10-11 wk	Rats--113-114 wk; mice--113 wk
Necropsy or Terminal-Kill Dates			
4/26/78	10/5/78-10/7/78	Rats--5/1/79-5/6/79; mice--5/1/79-5/4/79	Rats--12/4/81-12/11/81; mice--11/30/81-12/4/81
Method of Animal Distribution			
According to tables of random numbers	Assigned to cages according to one table of random numbers and then to groups according to another table of random numbers	Same as 14-d studies	Same as 14-d studies
Method of Animal Identification			
Ear punch	Ear punch	Ear punch	Ear punch
Feed			
Available ad libitum; Wayne Lab Blox® pellets (Allied Mills, Inc., Chicago, IL)	Same as single-administration studies	Same as single-administration studies	Available ad libitum; NIH 07 Open Formula (Zeigler Bros., Gardners, PA)
Bedding			
Heat-treated hardwood chips (Northeastern Products Corp., Warrensburg, NY) and sawdust (PWI, Inc., Lowville, NY)	Heat-treated hardwood chips (PWI, Inc., Lowville, NY)	Heat-treated hardwood chips (Northeastern Products, Warrensburg, NY)	Same as 13-wk studies
Water			
Available ad libitum; automatic watering system (Edstrom Industries, Waterford, WI)	Same as single-administration studies	Same as single-administration studies	Same as single-administration studies

TABLE 4. EXPERIMENTAL DESIGN AND MATERIALS AND METHODS IN THE GAVAGE STUDIES OF HC RED NO. 3 (Continued)

Single-Administration Studies	Fourteen-Day Studies	Thirteen-Week Studies	Two-Year Studies
ANIMALS AND ANIMAL MAINTENANCE (Continued)			
Cages			
Polycarbonate (Lab Products, Inc., Garfield, NJ)	Same as single-administration studies	Same as single-administration studies	Same as single-administration studies
Cage Filters			
Reemay spun-bonded polyester (Snow Filtration, Cincinnati, OH)	Same as single-administration studies	Same as single-administration studies	Same as single-administration studies
Animals per Cage			
5	5	5	5
Animal Room Environment			
Fluorescent light 12 h/d; 15 room air changes/h; rel hum--40%-60%; av temp--21°-23° C	Fluorescent light 12 h/d; 15 room air changes/h; rel hum--30%-50%; av temp--21°-23° C	Same as 14-d studies	Fluorescent light 12 h/d; rel hum--30%-50%, except 12/79--40%-60%; 15 room air changes/h; av temp--21°-23° C (a)
Other Chemicals on Test in Same Room			
None	None	None	None

(a) Ninety-seven percent of the temperature readings and 82% of the humidity readings were within the stated range.

observed for 3 weeks, and then assigned to cages according to a table of random numbers. The cages were then assigned to vehicle control and dosed groups according to another table of random numbers. Rats and mice were housed five per cage in polycarbonate cages. Wayne Lab Blox® pellets and water via an automatic watering system were available ad libitum. Further experimental details are summarized in Table 4.

Groups of 10 rats of each sex were administered HC Red No. 3 (0, 62, 125, 250, 500, or 1,000 mg/kg) in corn oil by gavage (13-gauge needle), 5 days per week for 13 weeks. Groups of 10 mice of each sex were administered 0, 15, 31, 62, 125, or 250 mg/kg on the same schedule (23-gauge needle). Animals were checked twice daily; moribund animals were killed. Individual animal weights were recorded weekly. At the end of the 13-week studies, survivors were killed. A necropsy was performed on all animals. Tissues and groups examined are listed in Table 4.

TWO-YEAR STUDIES

Study Design

Groups of 50 rats of each sex were administered 0, 250, or 500 mg/kg HC Red No. 3 in corn oil by gavage, 5 days per week for 105 weeks. Groups of 50 mice of each sex were administered 0, 125, or 250 mg/kg HC Red No. 3, 5 days per week for 104 weeks. Ten mice that were killed (gavage accidents) during the first month of the studies were replaced. The replacement animals were from pools of extra animals in each dose group that were specified for this purpose. Replacement animals were dosed during the first month of the studies.

Source and Specifications of Animals

The male and female F344/N rats and B6C3F$_1$ (C57BL/6N, female, × C3H/HeN MTV$^-$, male) mice used in these studies were produced under

strict barrier conditions at Charles River Breeding Laboratories under a contract to the Carcinogenesis Program. Breeding stocks for the foundation colony at the production facility originated at the National Institutes of Health Repository. Animals shipped for testing were progeny of defined microflora-associated parents that were transferred from isolators to barrier-maintained rooms. Animals were shipped to the testing laboratory at 4-5 weeks of age. The animals were quarantined at the testing facility for 20 days. Thereafter, a complete necropsy was performed on five animals of each sex and species to assess their health status. The rats were placed on study at 7-8 weeks of age and the mice at 8 weeks. The health of the animals was monitored during the course of the studies according to the protocols of the NTP Sentinel Animal Program (Appendix L).

A quality control skin grafting program has been in effect since early 1978 to monitor the genetic integrity of the inbred mice used to produce the hybrid $B6C3F_1$ test animal. In mid-1981, data were obtained that showed incompatibility between the NIH C3H reference colony and the C3H colony from a Program supplier. In August 1981, inbred parental lines of mice were further tested for genetic integrity via isozyme and protein electrophoretograms that demonstrate phenotype expressions of known genetic loci.

The C57BL/6 mice were homogeneous at all loci tested. Eighty-five percent of the C3H mice monitored were variant at one to three loci, indicating some heterogeneity in the C3H line from this supplier. Nevertheless, the genome of this line is more homogeneous than that of randomly bred stocks.

Male mice from the C3H colony and female mice from the C57BL/6 colony were used as parents for the hybrid $B6C3F_1$ mice used in these studies. The influence of the potential genetic nonuniformity in the hybrid mice on these results is not known, but results of the studies are not affected because concurrent controls were included in each study.

Animal Maintenance

Food and water were available ad libitum. Details of animal maintenance are summarized in Table 4.

Clinical Examinations and Pathology

All animals were observed twice daily, and clinical signs were recorded once per week. Body weights by cage were recorded once per week for the first 12 weeks of the studies and once per month thereafter. Mean body weights were calculated for each group. Moribund animals were killed, as were animals that survived to the end of the studies. A necropsy was performed on all animals, including those found dead unless they were excessively autolyzed or cannibalized. Thus, the number of animals from which particular organs or tissues were examined microscopically varies and is not necessarily equal to the number of animals that were placed on study in each group.

Examinations for grossly visible lesions were performed on major tissues or organs. Tissues were preserved in 10% neutral buffered formalin, embedded in paraffin, sectioned, and stained with hematoxylin and eosin. Tissues examined microscopically are listed in Table 4.

When the pathology examination was completed, the slides, individual animal data records, and summary tables were sent to an independent quality assurance laboratory. Individual animal records and tables were compared for accuracy, slides and tissue counts were verified, and histotechnique was evaluated. All tumor diagnoses, all target tissues, and all tissues from a randomly selected 10% of the animals were evaluated by a quality assurance pathologist. Slides of all target tissues and those about which the original and quality assurance pathologists disagreed were submitted to the Chairperson of the Pathology Working Group (PWG) for evaluation. Representative coded slides selected by the PWG Chairperson were reviewed by the PWG's pathologists, who reached a consensus and compared their findings with the

original and quality assurance diagnoses. When diagnostic differences were found, the PWG sent the appropriate slides and comments to the original pathologist for review. This procedure has been described, in part, by Maronpot and Boorman (1982) and Boorman et al. (1985). The final diagnosis represents a consensus of contractor pathologists and the NTP Pathology Working Group.

Nonneoplastic lesions are not examined routinely by the quality assurance pathologist or PWG. Certain nonneoplastic findings are reviewed by the quality assurance pathologist and PWG if they are considered part of the toxic response to a chemical or if they are deemed of special interest.

Statistical Methods

Data Recording: Data on this experiment were recorded in the Carcinogenesis Bioassay Data System (Linhart et al., 1974). The data elements include descriptive information on the chemicals, animals, experimental design, survival, body weight, and individual pathologic results, as recommended by the International Union Against Cancer (Berenblum, 1969).

Survival Analyses: The probability of survival was estimated by the product-limit procedure of Kaplan and Meier (1958) and is presented in the form of graphs. Animals were censored from the survival analyses at the time they were found dead of other than natural causes or were found to be missing; animals dying from natural causes were not 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) life table test for a dose-related trend. All reported P values for the survival analysis are two-sided.

Calculation of Incidence: The incidence of neoplastic or nonneoplastic lesions is given as the ratio of the number of animals bearing such lesions at a specific anatomic site to the number of animals in which that site was examined. In most instances, the denominators include only those animals for which the site was examined histologically. However, when macroscopic examination was required to detect lesions (e.g., skin or mammary tumors) prior to histologic sampling, or when lesions could have appeared at multiple sites (e.g., lymphomas), the denominators consist of the number of animals on which a necropsy was performed.

Analysis of Tumor Incidence: Three statistical methods are used to analyze tumor incidence data. The two that adjust for intercurrent mortality employ the classical method for combining contingency tables developed by Mantel and Haenszel (1959). Tests of significance included pairwise comparisons of high dose and low dose groups with vehicle controls and tests for overall dose-response trends.

For studies in which compound administration has little effect on survival, the results of the three alternative analyses will generally be similar. When differing results are obtained by the three methods, the final interpretation of the data will depend on the extent to which the tumor under consideration is regarded as being the cause of death. All reported P values for tumor analyses are one-sided.

Life Table Analyses--The first method of analysis assumed that all tumors of a given type observed in animals dying before the end of the studies were "fatal"; i.e., they either directly or indirectly caused the death of the animal. According to this approach, the proportions of tumor-bearing animals in the dosed and vehicle control groups were compared at each point in time at which an animal died with a tumor of interest. The denominators of these proportions were the total number of animals at risk in each group. These results, including the data from animals killed at the end of the studies, were then combined by the Mantel-Haenszel method to obtain an overall P value. This method of adjusting for intercurrent mortality is the life table method of Cox (1972) and of Tarone (1975).

Incidental Tumor Analyses--The second method of analysis assumed that all tumors of a given type observed in animals that died before the end of the studies were "incidental"; i.e., they were merely observed at necropsy in animals dying of an unrelated cause. According to this approach, the proportions of tumor-bearing animals in dosed and vehicle control groups were

compared in each of five time intervals: weeks 0-52, weeks 53-78, weeks 79-92, week 93 to the week before the terminal-kill period, and the terminal-kill period. The denominators of these proportions were the number of animals on which a necropsy was actually performed during the time interval. The individual time interval comparisons were then combined by the previously described method to obtain a single overall result. (See Haseman, 1984, for the computational details of both methods.)

Unadjusted Analyses--Primarily, survival-adjusted methods are used to evaluate tumor incidence. In addition, the results of the Fisher exact test for pairwise comparisons and the Cochran-Armitage linear trend test (Armitage, 1971; Gart et al., 1979) are given in the appendix containing the analyses of primary tumor incidence. These two tests are based on the overall proportion of tumor-bearing animals and do not adjust for survival differences.

Historical Control Data: Although the concurrent vehicle control group is always the first and most appropriate vehicle control group used for making decisions, there are certain instances in which historical control data can be helpful in the overall evaluation of tumor incidence. Consequently, control tumor incidences from the NTP historical control data base (Haseman et al., 1984) are included for those tumors in these studies appearing to show compound-related effects.

III. RESULTS

RATS

SINGLE-ADMINISTRATION STUDIES

FOURTEEN-DAY STUDIES

THIRTEEN-WEEK STUDIES

TWO-YEAR STUDIES

Body Weights and Clinical Signs

Survival

Pathology and Statistical Analyses of Results

MICE

SINGLE-ADMINISTRATION STUDIES

FOURTEEN-DAY STUDIES

THIRTEEN-WEEK STUDIES

TWO-YEAR STUDIES

Body Weights and Clinical Signs

Survival

Pathology and Statistical Analyses of Results

SINGLE-ADMINISTRATION STUDIES

None of the rats died before the end of the studies (Table 5). Differences in mean body weight gains were not dose related. The urine of all dosed rats was orange to red for 1 day after the animals were dosed.

TABLE 5. SURVIVAL AND MEAN BODY WEIGHTS OF RATS IN THE SINGLE-ADMINISTRATION GAVAGE STUDIES OF HC RED NO. 3

Dose (mg/kg)	Survival (a)	Mean Body Weights (grams) Initial	Final	Change (b)
MALE				
62	5/5	114 ± 2	180 ± 3	+ 66 ± 3
125	5/5	110 ± 5	176 ± 8	+ 66 ± 4
250	5/5	111 ± 3	183 ± 5	+ 72 ± 3
500	5/5	111 ± 3	180 ± 5	+ 69 ± 3
1,000	5/5	99 ± 4	168 ± 8	+ 69 ± 4
FEMALE				
62	5/5	88 ± 3	123 ± 5	+ 35 ± 3
125	5/5	82 ± 5	118 ± 5	+ 36 ± 2
250	5/5	93 ± 3	132 ± 5	+ 39 ± 2
500	5/5	85 ± 3	119 ± 6	+ 34 ± 4
1,000	5/5	83 ± 2	116 ± 2	+ 33 ± 1

(a) Number surviving/number initially in group
(b) Mean body weight change of the group ± standard error of the mean

FOURTEEN-DAY STUDIES

None of the rats died before the end of the studies (Table 6). The urine of all dosed animals was maroon to orange throughout the studies. Differences in mean body weight gains were not dose related. Dark thyroid glands were observed in 5/5 male rats that received 1,000 mg/kg, 2/5 males that received 500 mg/kg, and 2/5 males that received 250 mg/kg.

TABLE 6. SURVIVAL AND MEAN BODY WEIGHTS OF RATS IN THE FOURTEEN-DAY GAVAGE STUDIES OF HC RED NO. 3

Dose (mg/kg)	Survival (a)	Mean Body Weights (grams)			Final Weight Relative to Vehicle Controls (percent)
		Initial	Final	Change (b)	
MALE					
0	5/5	102 ± 3	167 ± 6	+ 65 ± 4	--
62	5/5	100 ± 6	151 ± 11	+ 51 ± 5	91.4
125	5/5	108 ± 4	182 ± 7	+ 74 ± 5	109.0
250	5/5	96 ± 3	148 ± 5	+ 52 ± 2	88.6
500	5/5	100 ± 2	165 ± 2	+ 65 ± 4	98.8
1,000	5/5	100 ± 4	159 ± 4	+ 59 ± 3	95.2
FEMALE					
0	5/5	87 ± 3	114 ± 5	+ 27 ± 2	--
62	5/5	96 ± 3	125 ± 4	+ 29 ± 0	109.6
125	5/5	94 ± 4	124 ± 4	+ 30 ± 2	108.8
250	5/5	89 ± 4	119 ± 2	+ 30 ± 2	104.4
500	5/5	93 ± 3	123 ± 3	+ 30 ± 2	107.9
1,000	5/5	90 ± 3	118 ± 3	+ 28 ± 1	103.5

(a) Number surviving/number in group
(b) Mean body weight change of the group ± standard error of the mean

THIRTEEN-WEEK STUDIES

None of the rats died before the end of the studies (Table 7). Final mean body weights relative to vehicle controls were 7% lower for male rats that received 1,000 mg/kg and 5% lower for the male rat group that received 500 mg/kg. Final mean body weights of dosed female rats were greater than those of the vehicle controls. The urine of dosed animals was orange to purple throughout the studies. Granules of a brown to golden-brown pigment were found in the cytoplasm of the thyroid gland follicular epithelial cells in 10/10 males and 10/10 females that received 1,000 mg/kg and 10/10 males and 7/10 females that received 500 mg/kg but not in any of the rats that received 250 mg/kg. Similar pigment was found in the cytoplasm of convoluted tubular epithelial cells in the kidneys of all rats that received 1,000 mg/kg, in 7/10 males and 10/10 females that received 500 mg/kg, and in 6/10 males and 7/10 females that received 250 mg/kg. No other microscopic observations attributable to HC Red No. 3 administration were noted.

Dose Selection Rationale: The doses selected for rats for the 2-year studies were 250 and 500 mg/kg HC Red No. 3. These doses were selected because of the intense pigmentation in the thyroid gland and kidneys at the 1,000 mg/kg dose. Although documented functional changes that can be related to the presence of pigment as seen in these studies have not been found, there was concern that potentially life-threatening functional changes might eventually be produced in the 1,000 mg/kg dose groups during the 2-year studies.

TABLE 7. SURVIVAL AND MEAN BODY WEIGHTS OF RATS IN THE THIRTEEN-WEEK GAVAGE STUDIES OF HC RED NO. 3

Dose (mg/kg)	Survival (a)	Mean Body Weights (grams) Initial	Final	Change (b)	Final Weight Relative to Vehicle Controls (percent)
MALE					
0	10/10	164 ± 2	357 ± 5	+ 193 ± 5	--
62	10/10	159 ± 4	352 ± 4	+ 193 ± 4	98.5
125	10/10	161 ± 4	350 ± 6	+ 189 ± 4	98.0
250	10/10	160 ± 3	356 ± 6	+ 196 ± 5	99.6
500	10/10	154 ± 5	338 ± 6	+ 184 ± 4	94.7
1,000	10/10	157 ± 4	333 ± 6	+ 176 ± 3	93.3
FEMALE					
0	10/10	121 ± 2	196 ± 2	+ 75 ± 2	--
62	10/10	118 ± 3	199 ± 3	+ 81 ± 2	101.6
125	10/10	119 ± 2	202 ± 2	+ 83 ± 3	102.9
250	10/10	122 ± 2	205 ± 3	+ 83 ± 1	104.8
500	10/10	123 ± 2	202 ± 4	+ 79 ± 2	102.9
1,000	10/10	117 ± 3	198 ± 3	+ 81 ± 2	101.0

(a) Number surviving/number in group
(b) Mean body weight change of the group ± standard error of the mean

TWO-YEAR STUDIES

Body Weights and Clinical Signs

Throughout most of the study, there was little or no difference in the mean body weights of dosed rats of either sex as compared with those of vehicle controls (Table 8 and Figure 1). No compound-related clinical signs were observed.

TABLE 8. MEAN BODY WEIGHTS AND SURVIVAL OF RATS IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

Weeks on Study	Vehicle Control		250 mg/kg			500 mg/kg		
	Av. Wt. (grams)	No. of Survivors	Av. Wt. (grams)	Wt. (percent of veh controls)	No. of Survivors	Av. Wt. (grams)	Wt. (percent of veh controls)	No. of Survivors
MALE								
0	153	50	152	99	50	150	98	50
1	205	50	202	99	50	200	98	50
2	232	50	229	99	50	226	97	50
3	254	50	249	98	50	246	97	50
4	270	50	269	100	50	265	98	50
5	285	50	284	100	50	279	98	50
6	299	50	299	100	50	291	97	50
7	312	50	312	100	50	300	96	50
8	320	50	321	100	50	313	98	50
9	329	50	330	100	50	323	98	50
10	339	50	339	100	50	331	98	50
11	347	50	347	100	50	340	98	50
12	355	50	355	100	50	348	98	50
15	375	50	375	100	50	368	98	50
19	397	50	394	99	50	389	98	50
23	413	50	418	101	50	407	99	50
28	431	50	433	100	50	422	98	50
33	433	50	436	101	50	429	99	50
38	453	50	455	100	50	445	98	50
42	471	50	467	99	50	459	97	50
46	481	50	481	100	49	470	98	50
50	487	50	488	100	49	475	98	49
54	493	49	493	100	49	477	97	49
59	502	49	502	100	49	485	97	48
63	503	49	503	100	49	492	98	47
67	511	49	507	99	49	495	97	45
72	512	48	501	98	48	493	96	44
77	508	48	509	100	45	495	97	41
81	506	46	506	100	45	493	97	37
85	504	45	507	101	45	496	98	37
90	502	41	503	100	44	496	99	35
94	495	41	499	101	41	485	98	34
99	481	39	498	104	37	485	101	33
104	479	34	496	104	34	477	100	32
105	473	34	491	104	33	471	100	32
FEMALE								
0	114	50	116	102	50	113	99	50
1	141	50	142	101	50	137	97	50
2	152	50	153	101	50	149	98	50
3	161	50	161	100	50	155	96	50
4	168	50	168	100	50	164	98	50
5	174	50	176	101	50	170	98	50
6	179	50	180	101	50	174	97	50
7	184	50	184	100	50	179	97	50
8	187	50	187	100	50	182	97	50
9	189	50	191	101	50	186	98	50
10	192	50	192	100	50	189	98	50
11	194	50	195	101	50	190	98	50
12	196	50	198	101	50	194	99	50
15	204	50	203	100	50	199	98	50
19	211	50	211	100	50	204	97	50
23	218	50	218	100	50	213	98	50
28	224	50	223	100	50	217	97	50
33	228	50	228	100	50	220	96	50
38	235	50	235	100	50	224	95	50
42	243	50	244	100	50	234	96	50
46	249	50	252	101	50	241	97	50
50	254	50	254	100	50	244	96	50
54	260	50	259	100	50	250	96	50
59	271	50	270	100	50	258	95	50
63	278	50	278	100	49	267	96	50
67	285	50	286	100	48	270	95	50
72	294	50	297	101	48	279	95	49
77	309	50	308	100	48	288	93	49
81	308	50	313	102	47	293	95	46
85	313	49	325	104	46	300	96	44
90	321	44	327	102	45	303	94	42
94	320	44	329	103	42	302	94	42
99	322	42	331	103	41	309	96	36
104	320	40	334	104	39	313	98	34
105	317	39	332	105	38	314	99	34

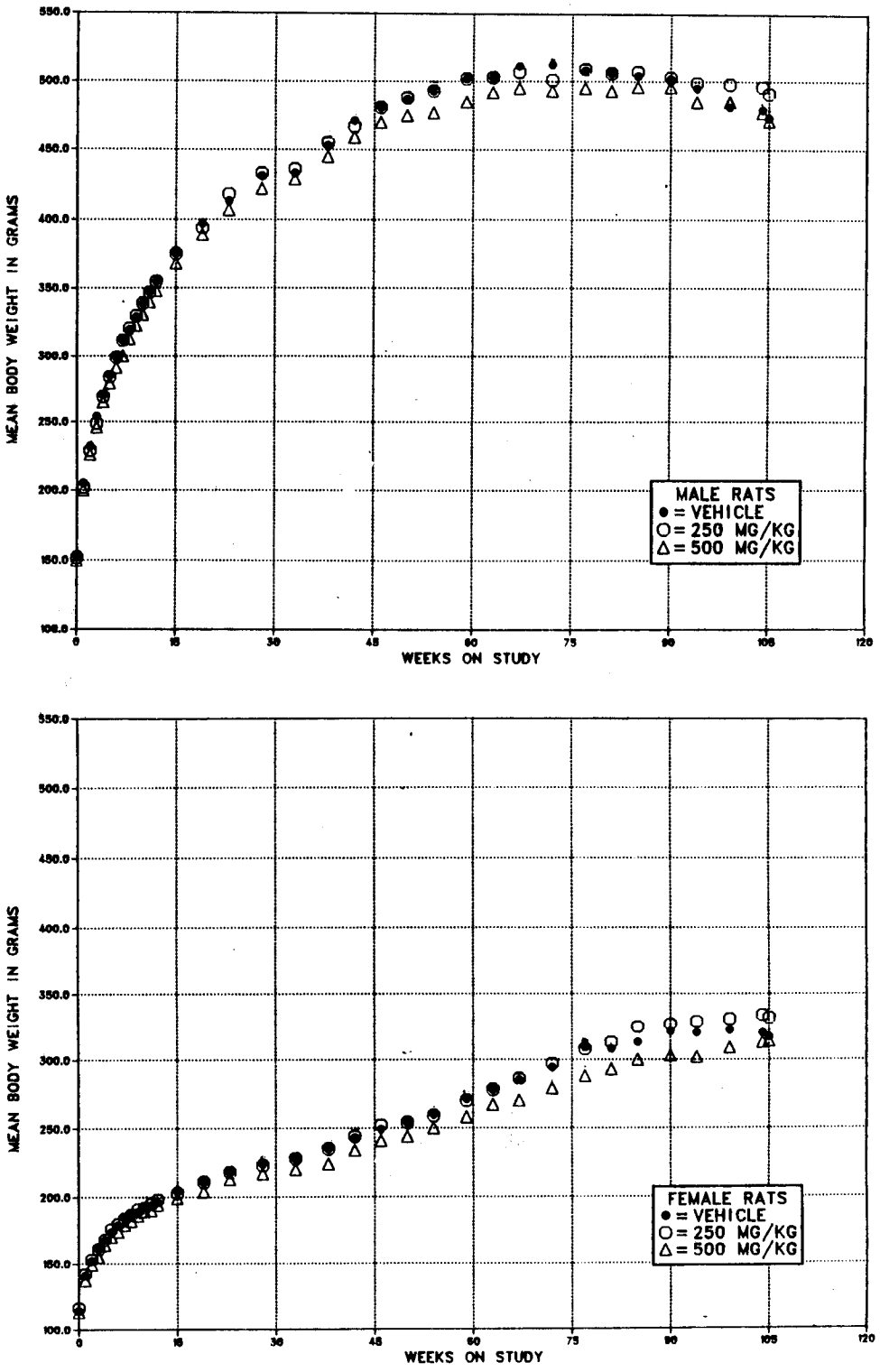


FIGURE 1. GROWTH CURVES FOR RATS ADMINISTERED HC RED NO. 3 IN CORN OIL BY GAVAGE FOR TWO YEARS

Survival

Estimates of the probabilities of the survival of male and female rats administered HC Red No. 3 by gavage at the doses used in these studies and those of the vehicle controls are shown in the Kaplan and Meier curves in Figure 2. No significant differences in survival were observed between any groups of either sex (Table 9).

Pathology and Statistical Analyses of Results

This section describes significant or noteworthy changes in the incidence of rats with neoplastic or nonneoplastic lesions of the urinary bladder, kidney, mammary gland, multiple organs, seminal vesicles, adrenal gland, thyroid gland, uterus, hematopoietic system, and eyes. Histopathologic findings on neoplasms in rats are summarized in Appendix A (Tables A1 and A2); Appendix A (Tables A3 and A4) also gives the survival and tumor status for individual male and female rats. Findings on nonneoplastic lesions are summarized in Appendix C (Tables C1 and C2). Appendix E (Tables E1 and E2) contains the statistical analyses of those primary tumors that occurred with an incidence of at least 5% in one of the three groups. The statistical analyses used are discussed in Chapter II (Statistical Methods) and Appendix E (footnotes). Historical incidences of tumors in corn oil vehicle control animals are listed in Appendix F.

TABLE 9. SURVIVAL OF RATS IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

	Vehicle Control	250 mg/kg	500 mg/kg
MALE (a)			
Animals initially in study	50	50	50
Nonaccidental deaths before termination (b)	15	14	14
Accidental deaths	1	2	4
Killed at termination	33	33	32
Died during termination period	1	1	0
Survival P values (c)	0.924	0.875	0.987
FEMALE (a)			
Animals initially in study	50	50	50
Nonaccidental deaths before termination (b)	11	11	16
Accidental deaths	0	1	0
Killed at termination	39	38	34
Survival P values (c)	0.266	0.954	0.314

(a) Terminal kill period: male--weeks 105-106; female--weeks 106-107
(b) Includes animals killed in a moribund condition
(c) The result of the life table trend test is in the vehicle control column, and the results of the life table pairwise comparisons with the vehicle controls are in the dosed columns.

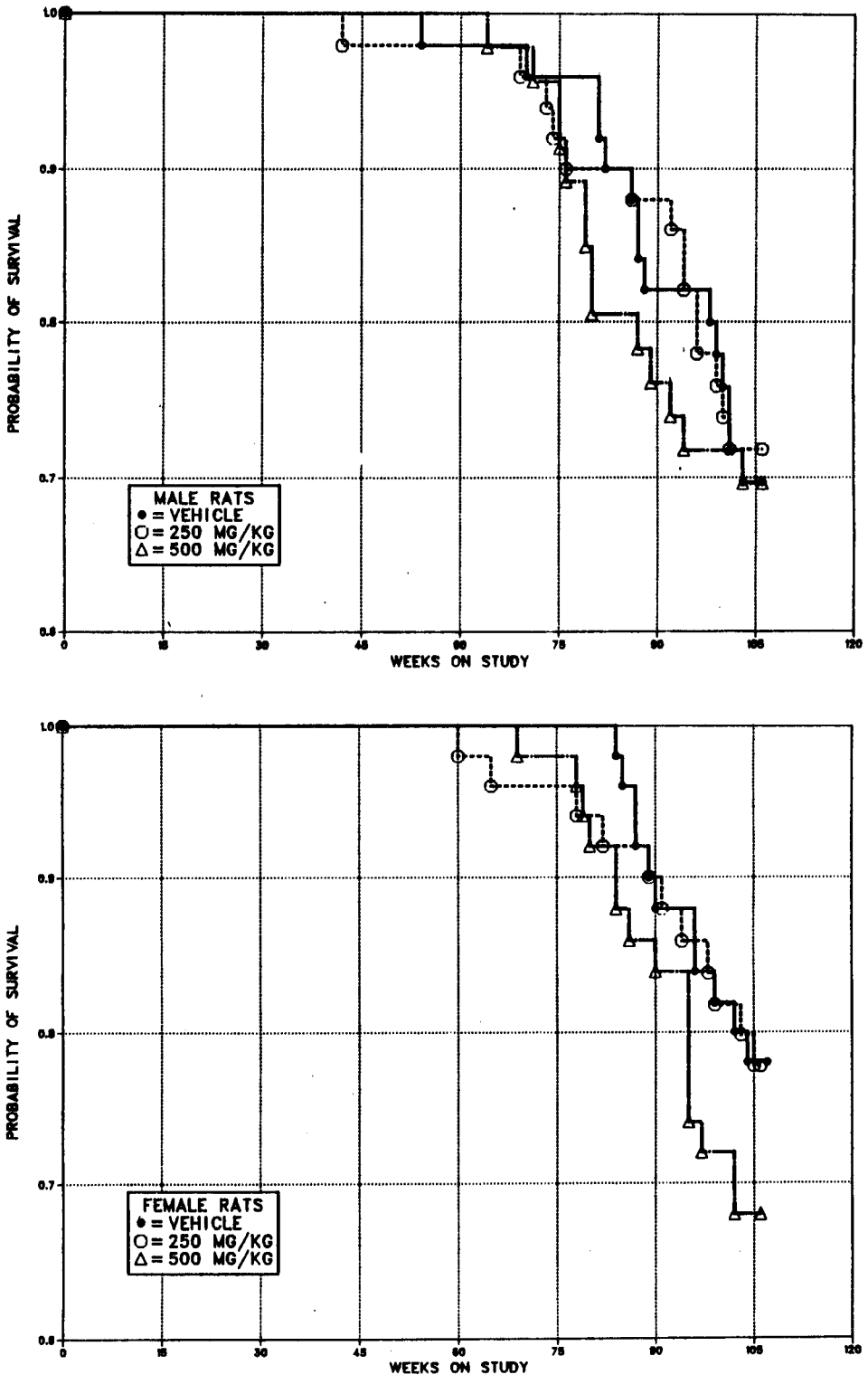


FIGURE 2. KAPLAN-MEIER SURVIVAL CURVES FOR RATS ADMINISTERED HC RED NO. 3 IN CORN OIL BY GAVAGE FOR TWO YEARS

Urinary System

Urinary Bladder--Transitional cell papillomas were observed in one high dose male rat, two low dose female rats, and one high dose female rat (Appendix A, Tables A3 and A4, and Appendix F, Table F1). These tumors were generally characterized as papillary projections into the bladder lumen and consisted of a fibrocapsular core covered by multilayers of well-differentiated transitional cell epithelium. The lesion in one rat tended to be more solid and flat and consisted of transitional cell epithelium.

Kidney--Nephropathy was observed at increased incidences in dosed female rats (vehicle control, 7/50; low dose, 12/50; high dose, 20/50). The kidneys of the female rats were reviewed in a blind fashion and graded for severity of nephropathy. The increased incidence was confirmed but was due to very minimal lesions. The average degree of severity actually decreased with dose.

Mammary Gland: The incidences of fibroadenomas and of fibroadenomas or cystadenomas (combined) in low dose (but not high dose) female rats were significantly greater than those in the vehicle controls (Table 10).

Multiple Organs: Pigmentation of the kidney tubules, thyroid gland, or multiple organs was observed in 0/50 vehicle control males, 49/50 low dose males, 49/50 high dose males, 0/50 vehicle control females, 50/50 low dose females, and 48/50 high dose females.

Seminal Vesicle: Atrophy of the seminal vesicles was observed at increased incidence in dosed male rats (vehicle control, 1/50, 2%; low dose, 12/50, 24%; high dose, 13/50, 26%). The lesions diagnosed as atrophy were very minimal, often involving a few flattened cells. The seminal vesicles in the male rats were reexamined. The reevaluation of the tissues confirmed that the changes were minimal and subtle. Further, the reviewing pathologist found greater incidences of the change in all groups than did the original pathologist and no difference in the incidences between vehicle control and dosed groups. It was concluded that this is a normal change in aging F344/N male rats and was not a compound-related effect.

TABLE 10. ANALYSIS OF MAMMARY GLAND TUMORS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (a)

	Vehicle Control	250 mg/kg	500 mg/kg
Fibroadenoma (b)			
Overall Rates	14/50 (28%)	24/50 (48%)	11/50 (22%)
Adjusted Rates	34.8%	55.5%	30.1%
Terminal Rates	13/39 (33%)	19/38 (50%)	9/34 (26%)
Life Table Tests	P=0.469N	P=0.029	P=0.465N
Incidental Tumor Tests	P=0.389N	P=0.019	P=0.433N
Cystadenoma			
Overall Rates	0/50 (0%)	1/50 (2%)	0/50 (0%)
Adenocarcinoma			
Overall Rates	0/50 (0%)	1/50 (2%)	2/50 (4%)
Cystadenoma or Fibroadenoma			
Overall Rates	14/50 (28%)	25/50 (50%)	11/50 (22%)
Adjusted Rates	34.8%	56.6%	30.1%
Terminal Rates	13/39 (33%)	19/38 (50%)	9/34 (26%)
Life Table Tests	P=0.470N	P=0.020	P=0.465N
Incidental Tumor Tests	P=0.376N	P=0.012	P=0.433N

(a) The statistical analyses used are discussed in Chapter II (Statistical Methods) and Appendix E (footnotes).
(b) Historical incidence at testing laboratory (mean ± SD): 80/300, 27% ± 6%; historical incidence in NTP studies: 269/1,147, 23% ± 9%

Negative Trends: The following statistically significant ($P<0.05$) negative trends and/or decreasing incidences of tumors were detected in dosed rats:

- Adrenal gland pheochromocytoma or pheochromocytoma, malignant (combined) in male rats (vehicle control, 20/49, 40%; low dose, 13/50, 26%; high dose, 11/50, 22%)
- Thyroid gland C-cell carcinoma and adenoma or carcinoma (combined) in male rats (vehicle control, 12/49, 24%; low dose, 5/49, 10%; high dose, 4/50, 8%)
- Uterine endometrial stromal sarcoma in female rats (vehicle control, 3/50, 6%; low dose, 0/50; high dose, 0/50)
- Mononuclear cell leukemia in both sexes (male: vehicle control, 9/50, 18%; low dose, 3/50, 6%; high dose, 3/50, 6%; female: vehicle control, 10/50, 20%; low dose, 6/50, 12%; high dose, 3/50, 6%)
- Mammary gland fibroadenomas in male rats (vehicle control, 8/50, 16%; low dose, 2/50, 4%; high dose, 2/50, 4%)

Eyes: Retinopathy and cataracts were observed at increased incidences in high dose male and low dose female rats (Table 11). These lesions have been reported in rats housed in the top row of cages in earlier studies at this laboratory (HC Blue No. 1 and HC Blue No. 2) and are believed to be related to cage placement relative to the light source rather than to test compound administration. These studies were conducted before cages were routinely rotated during 2-year studies. Therefore, the rats remained in their initially assigned cage positions for the full length of the studies. High dose males and low dose females (the groups affected) were housed in the uppermost rows (closest to the light source) of their respective cage racks. The remaining groups, which were much less affected, were housed farther from the light source (Table 11).

TABLE 11. INCIDENCE OF CATARACTS AND RETINOPATHY IN RATS IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

	Male			Female		
	Vehicle Control	250 mg/kg	500 mg/kg	Vehicle Control	250 mg/kg	500 mg/kg
Number of rats examined	50	50	50	50	50	50
Cataracts	2	0	19	1	19	0
Retinopathy	2	0	20	1	19	0

SINGLE-ADMINISTRATION STUDIES

None of the mice died before the end of the studies (Table 12). Changes in mean body weight gain were not dose related. The urine of all dosed animals was orange to red for 1 day after the animals were dosed.

TABLE 12. SURVIVAL AND MEAN BODY WEIGHTS OF MICE IN THE SINGLE-ADMINISTRATION GAVAGE STUDIES OF HC RED NO. 3

Dose (mg/kg)	Survival (a)	Mean Body Weights (grams) Initial	Final	Change (b)
MALE				
31	5/5	23.2 ± 0.2	26.6 ± 0.7	+ 3.4 ± 0.7
62	5/5	22.8 ± 1.0	26.6 ± 1.0	+ 3.8 ± 0.4
125	5/5	23.0 ± 0.5	26.2 ± 0.6	+ 3.2 ± 0.8
250	5/5	23.0 ± 0.7	26.4 ± 0.7	+ 3.4 ± 0.7
500	5/5	23.0 ± 0.5	25.6 ± 0.7	+ 2.6 ± 0.4
FEMALE				
31	5/5	17.2 ± 0.6	19.6 ± 0.7	+ 2.4 ± 0.2
62	5/5	18.6 ± 0.4	21.2 ± 0.7	+ 2.6 ± 0.4
125	5/5	18.0 ± 0.4	21.0 ± 0.3	+ 3.0 ± 0.3
250	5/5	18.2 ± 0.2	20.6 ± 0.2	+ 2.4 ± 0.2
500	5/5	18.2 ± 0.5	20.2 ± 0.6	+ 2.0 ± 0.3

(a) Number surviving/number initially in group
(b) Mean body weight change of the group ± standard error of the mean

FOURTEEN-DAY STUDIES

None of the mice died before the end of the studies (Table 13). The urine of all dosed animals was maroon to orange throughout the studies.

Mean body weight gains by dosed and vehicle control groups were comparable.

TABLE 13. SURVIVAL AND MEAN BODY WEIGHTS OF MICE IN THE FOURTEEN-DAY GAVAGE STUDIES OF HC RED NO. 3

Dose (mg/kg)	Survival (a)	Mean Body Weights (grams) Initial	Final	Change (b)	Final Weight Relative to Vehicle Controls (percent)
MALE					
0	5/5	20.4 ± 1.1	26.0 ± 0.9	+5.6 ± 0.2	--
31	5/5	21.4 ± 0.4	24.8 ± 0.7	+3.4 ± 0.4	95.4
62	5/5	20.0 ± 0.8	23.6 ± 0.7	+3.6 ± 0.4	90.8
125	5/5	22.2 ± 0.5	26.0 ± 0.5	+3.8 ± 0.4	100.0
250	5/5	21.2 ± 0.9	24.8 ± 0.6	+3.6 ± 0.9	95.4
500	5/5	19.6 ± 0.5	25.2 ± 0.4	+5.6 ± 0.6	96.9
FEMALE					
0	5/5	16.0 ± 0.8	19.0 ± 0.4	+3.0 ± 0.4	--
31	5/5	16.4 ± 0.5	18.8 ± 0.7	+2.4 ± 0.4	98.9
62	5/5	15.6 ± 0.4	18.8 ± 0.6	+3.2 ± 0.4	98.9
125	5/5	15.8 ± 0.4	19.0 ± 0.5	+3.2 ± 0.7	100.0
250	5/5	16.0 ± 0.4	19.6 ± 0.2	+3.6 ± 0.4	103.2
500	5/5	14.4 ± 0.4	18.0 ± 0.8	+3.6 ± 1.2	94.7

(a) Number surviving/number initially in group
(b) Mean body weight change of the group ± standard error of the mean

THIRTEEN-WEEK STUDIES

All deaths that occurred were related to gavage technique (Table 14). The final mean body weight of male mice administered 250 mg/kg was 7% lower than that of the vehicle controls. All dosed animals had red urine throughout the studies. No compound-related gross or microscopic pathologic effects were observed.

Although 500 mg/kg HC Red No. 3 was administered with no effect in the 14-day studies, the highest dose used in the 13-week studies was 250 mg/kg because at higher concentrations the viscosity of HC Red No. 3 made precise administration difficult (see *Dose Selection Rationale*).

Dose Selection Rationale: The highest dose of HC Red No. 3 in mice was limited by the viscosity of the corn oil suspension and by the diameter of the gavaging needle rather than by toxicity. During both the single-administration and the 14-day studies, the highest dose was 500 mg/kg, administered in a dose volume of 20 ml/kg, and it produced no effect. The largest gavage volume used in NTP 2-year studies is 10 ml/kg, which would have required a suspension concentration of 50 mg/ml in order to deliver 500 mg/kg. At this concentration, the suspension was extremely difficult to draw through the gavage needle and the 25 mg/ml suspension was the maximum dose that could be gavaged.

TABLE 14. SURVIVAL AND MEAN BODY WEIGHTS OF MICE IN THE THIRTEEN-WEEK GAVAGE STUDIES OF HC RED NO. 3

Dose (mg/kg)	Survival (a)	Mean Body Weights (grams) Initial (b)	Final	Change (c)	Final Weight Relative to Vehicle Controls (percent)
MALE					
0	8/10	26.4 ± 0.5	39.0 ± 0.9	+ 12.2 ± 0.9	--
15	7/10	25.2 ± 0.4	39.0 ± 0.8	+ 13.7 ± 0.9	100.0
31	10/10	26.1 ± 0.4	39.0 ± 0.8	+ 12.9 ± 0.6	100.0
62	9/10	25.9 ± 0.6	39.7 ± 0.9	+ 14.1 ± 0.6	101.8
125	7/10	25.8 ± 0.4	39.3 ± 0.4	+ 13.4 ± 0.4	100.8
250	9/10	25.3 ± 0.6	36.4 ± 0.8	+ 11.3 ± 0.5	93.3
FEMALE					
0	9/10	19.1 ± 0.3	29.0 ± 0.8	+ 9.9 ± 0.6	--
15	10/10	20.2 ± 0.4	30.4 ± 1.3	+ 10.2 ± 1.0	104.8
31	9/10	19.4 ± 0.3	29.9 ± 1.1	+ 10.6 ± 0.8	103.1
62	9/10	19.4 ± 0.3	28.6 ± 0.3	+ 9.4 ± 0.4	98.6
125	9/10	19.4 ± 0.4	29.9 ± 0.9	+ 10.6 ± 0.7	103.1
250	10/10	19.7 ± 0.3	28.8 ± 0.6	+ 9.1 ± 0.3	99.3

(a) Number surviving/number initially in group. All deaths were due to gavage technique rather than to toxicity.
(b) Initial mean body weight ± standard error of the mean of all animals in the group. Subsequent calculations are based on those animals surviving to the end of the study.
(c) Mean body weight change of the survivors ± standard error of the mean

TWO-YEAR STUDIES

Body Weights and Clinical Signs

Mean body weights of high dose mice of each sex were comparable to or greater than those of the vehicle controls throughout most of the study (Table 15 and Figure 3). The body weights of both male and female low dose mice tended to be lower than those of other groups throughout the study. No compound-related clinical signs were observed

TABLE 15. MEAN BODY WEIGHTS AND SURVIVAL OF MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

Weeks on Study	Vehicle Control		125 mg/kg			250 mg/kg		
	Av. Wt. (grams)	No. of Survivors	Av. Wt. (grams)	Wt. (percent of veh controls)	No. of Survivors	Av. Wt. (grams)	Wt. (percent of veh controls)	No. of Survivors
MALE								
0	25.6	50	26.2	102	50	26.7	104	50
1	28.8	50	28.3	98	50	29.1	101	50
2	29.6	50	28.5	96	50	28.3	96	50
3	30.9	50	29.5	95	50	30.3	98	50
4	31.6	50	30.9	98	50	32.1	102	50
5	33.2	50	31.2	94	50	32.3	97	50
6	33.7	50	33.0	98	50	34.9	104	50
7	35.5	50	33.2	94	50	35.1	99	50
8	36.3	50	34.1	94	50	36.8	101	50
9	36.5	50	34.6	95	50	37.4	102	50
10	36.7	50	35.2	96	50	37.9	103	50
11	37.9	50	35.0	92	50	38.7	102	49
12	38.8	50	35.1	90	50	38.9	100	49
15	40.6	50	36.5	90	50	42.0	103	49
19	41.4	50	38.7	93	50	43.5	105	48
23	44.5	50	41.1	92	50	45.5	102	48
28	46.1	50	42.3	92	50	48.0	104	48
33	47.0	50	44.2	94	50	48.1	102	48
38	47.1	50	44.5	94	50	49.1	104	48
42	48.4	50	45.6	94	50	50.0	103	48
46	48.8	49	46.6	95	50	50.6	104	48
50	49.1	49	45.6	93	50	50.6	103	47
54	49.4	49	47.0	95	50	52.0	105	47
59	50.0	49	47.5	95	50	51.6	103	47
63	50.8	46	48.1	95	50	52.0	102	47
67	51.6	46	49.0	95	50	52.1	101	47
72	50.5	46	48.0	95	50	51.0	101	46
77	50.9	44	49.1	96	48	52.6	103	45
81	50.9	43	49.1	96	47	52.2	103	45
85	50.6	40	49.8	98	45	51.6	102	44
90	49.4	38	48.3	98	44	50.2	102	40
94	48.9	34	48.0	98	42	51.8	106	33
99	47.8	31	47.0	98	42	50.0	105	30
105	44.4	30	42.9	97	41	46.6	105	29
FEMALE								
0	18.9	50	19.1	101	50	19.3	102	50
1	20.8	50	20.2	97	50	21.3	102	50
2	21.9	50	21.4	98	50	21.5	98	50
3	23.1	50	21.8	94	50	23.0	100	50
4	23.7	50	23.0	97	50	23.2	98	49
5	24.5	50	23.3	95	50	24.6	100	49
6	24.5	50	24.4	100	50	25.4	104	49
7	25.5	50	25.3	99	50	25.3	99	49
8	25.6	50	24.7	96	50	26.1	102	49
9	26.0	50	25.0	96	50	26.4	102	49
10	26.0	50	26.2	101	50	26.7	103	49
11	26.6	50	25.6	96	50	27.5	103	49
12	27.1	50	26.6	98	50	27.4	101	49
15	28.1	50	26.5	94	49	28.8	102	49
19	28.5	50	25.8	91	49	28.8	101	49
23	30.2	50	29.1	96	49	30.6	101	49
28	30.8	50	28.9	94	49	32.6	106	49
33	32.1	50	31.1	97	49	34.3	107	48
38	32.8	50	31.5	96	49	34.9	106	48
42	34.0	50	31.2	92	49	35.8	105	48
46	35.5	49	33.9	95	49	37.5	106	48
50	36.1	49	33.9	94	48	38.8	107	46
54	37.3	48	35.3	95	48	40.2	108	45
59	39.0	45	36.3	93	48	40.7	104	42
63	39.3	43	37.1	94	47	41.6	106	39
67	39.6	42	38.4	97	45	42.2	107	38
72	40.1	31	38.3	96	39	44.0	110	32
77	41.5	28	40.3	97	36	45.7	110	26
81	42.4	25	40.0	94	28	45.9	108	23
85	44.2	21	40.5	92	23	46.7	106	20
90	44.9	18	38.9	87	15	45.6	102	15
94	45.5	14	38.7	85	14	43.4	95	15
99	44.2	13	37.0	84	11	45.0	102	11
105	43.0	12	34.1	79	8	42.1	98	9

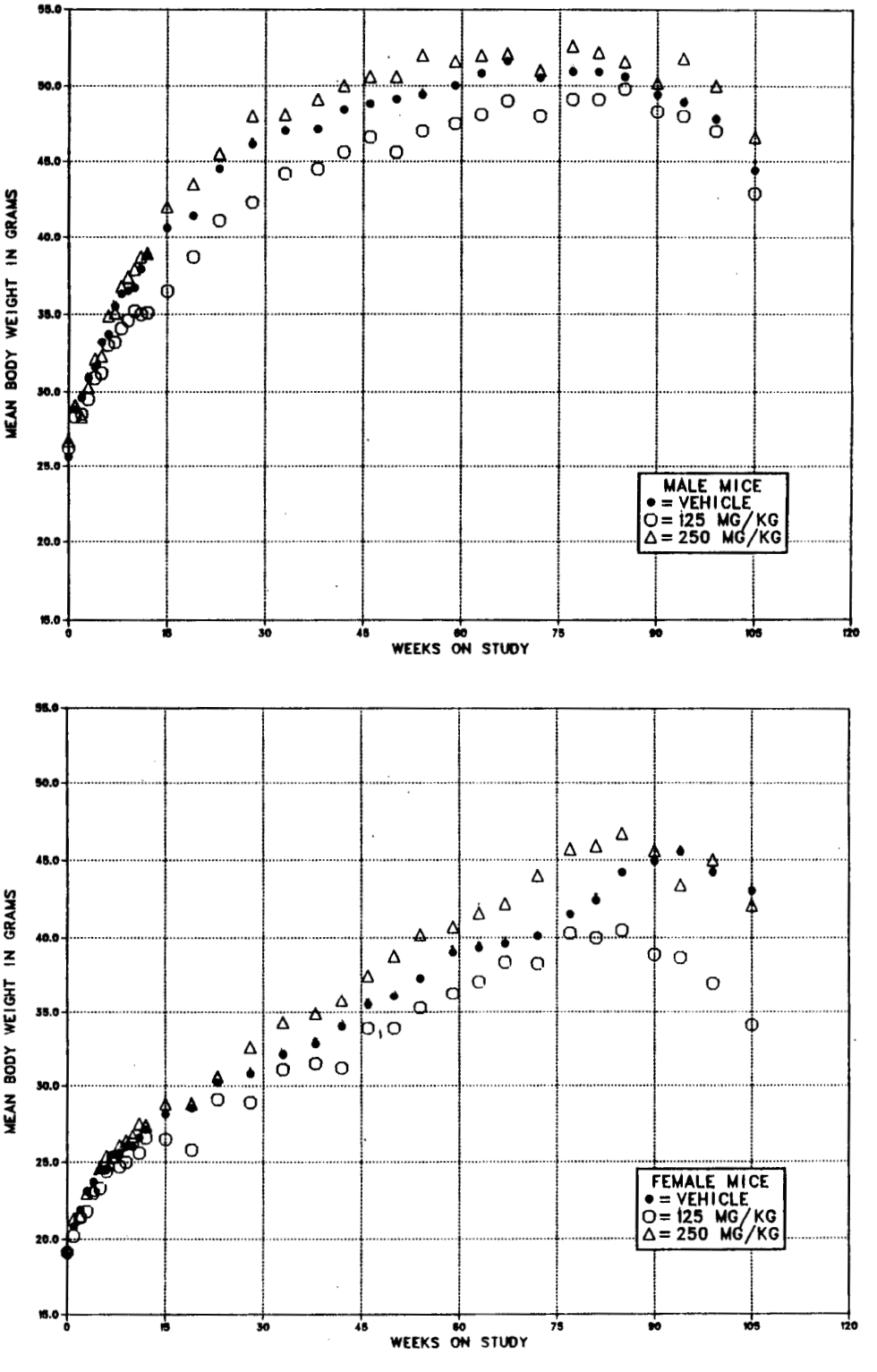


FIGURE 3. GROWTH CURVES FOR MICE ADMINISTERED HC RED NO. 3 IN CORN OIL BY GAVAGE FOR TWO YEARS

Survival

Estimates of the probabilities of survival of male and female mice administered HC Red No. 3 at the doses in these studies and those of the vehicle controls are shown in the Kaplan and Meier curves in Figure 4. In male mice, the survival of the low dose group was significantly greater ($P = 0.027$) than that of the vehicle control group (Table 16). The survival of all groups of female mice was unusually low in comparison with historical controls. This reduced survival was attributed to a reproductive tract infection that affected all groups of female mice.

Pathology and Statistical Analyses of Results

This section describes significant or noteworthy changes in the incidence of mice with neoplastic or nonneoplastic lesions of the liver, forestomach, thyroid gland, kidney, uterus, ovary, and multiple organs. Histopathologic findings on neoplasms in mice are summarized in Appendix B (Tables B1 and B2); Appendix B (Tables B3 and B4) also gives the survival and tumor status for individual male and female mice. Findings on nonneoplastic lesions are summarized in Appendix D (Tables D1 and D2). Appendix E (Tables E3 and E4) contains the statistical analyses of those primary tumors that occurred with an incidence of at least 5% in one of the three groups. The statistical analyses used are discussed in Chapter II (Statistical Methods) and Appendix E (footnotes). Historical incidences of tumors in corn oil vehicle control animals are listed in Appendix F.

TABLE 16. SURVIVAL OF MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

	Vehicle Control	125 mg/kg	250 mg/kg
MALE (a)			
Animals initially in study	50	50	50
Nonaccidental deaths before termination (b)	20	9	21
Killed at termination	30	41	29
Survival P values (c)	0.970	0.027	0.916
FEMALE (a)			
Animals initially in study	50	50	50
Nonaccidental deaths before termination (b)	38	42	41
Killed at termination	12	8	9
Survival P values (c)	0.523	0.972	0.580

(a) Terminal kill period: week 105
(b) Includes animals killed in a moribund condition
(c) The result of the life table trend test is in the vehicle control column, and the results of the life table pairwise comparisons with the vehicle controls are in the dosed columns.

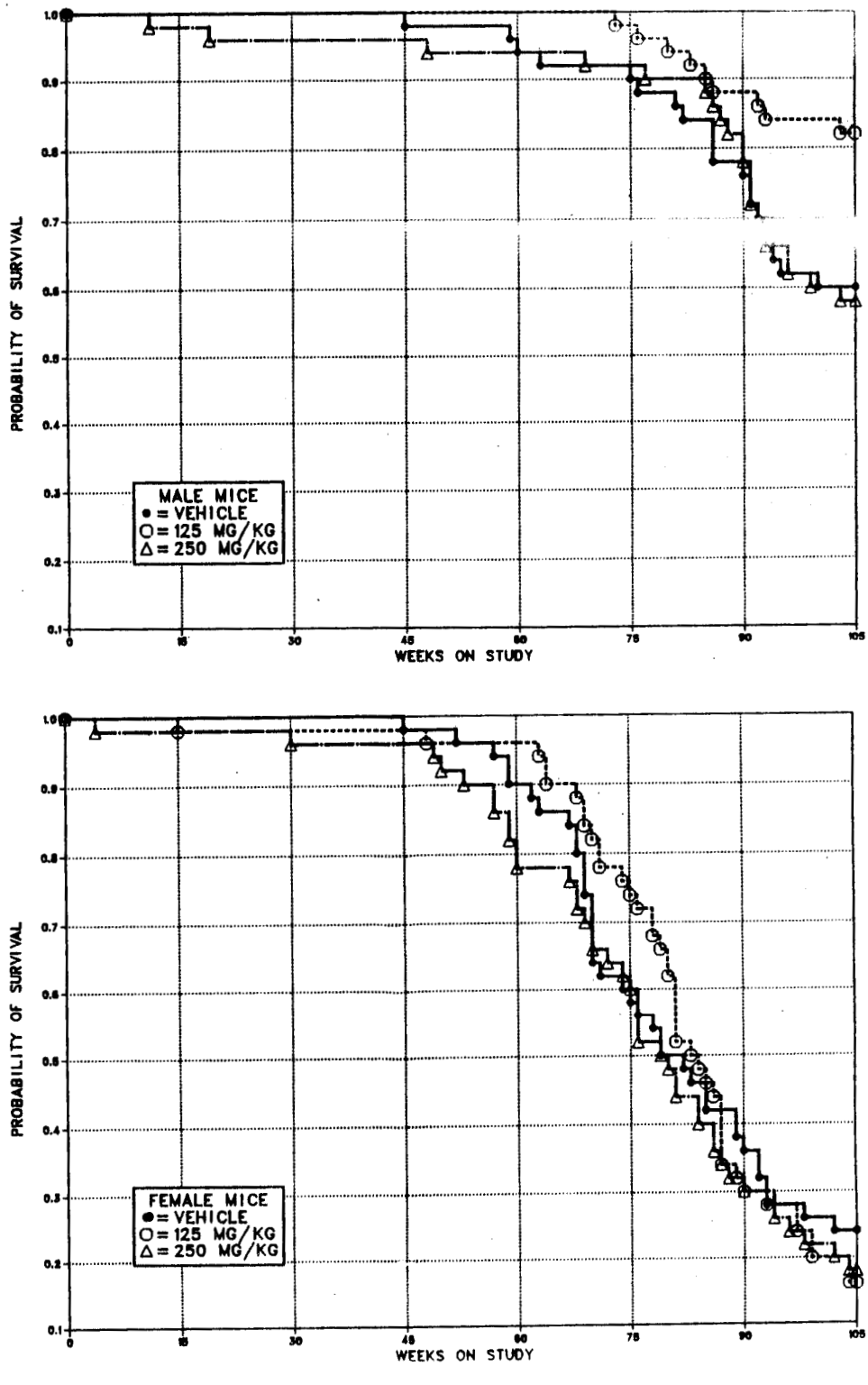


FIGURE 4. KAPLAN-MEIER SURVIVAL CURVES FOR MICE ADMINISTERED HC RED NO. 3 IN CORN OIL BY GAVAGE FOR TWO YEARS

Liver: Hepatocellular adenomas or carcinomas (combined) in male mice occurred with a significant positive trend, and the incidence of hepatocellular adenomas or carcinomas (combined) in high dose male mice was significantly greater than that in the vehicle controls (Table 17). The incidences of hepatocellular adenomas, hepatocellular carcinomas, and hepatocellular adenomas or carcinomas (combined) in low dose male mice were significantly lower than those in the vehicle controls (life table analysis). Hepatocellular adenomas in female mice occurred with a significant negative trend; no significant differences were observed for adenomas or carcinomas (combined). The hepatocellular carcinomas metastasized to the lung in three vehicle control males, one low dose male, and five high dose males but in none of the females.

TABLE 17. ANALYSIS OF LIVER TUMORS IN MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3 (a)

	Vehicle Control	125 mg/kg	250 mg/kg
MALE			
Hepatocellular Adenoma			
Overall Rates	11/50 (22%)	6/50 (12%)	16/50 (32%)
Adjusted Rates	33.5%	13.9%	49.4%
Terminal Rates	9/30 (30%)	4/41 (10%)	13/29 (45%)
Life Table Tests	P=0.118	P=0.048N	P=0.162
Incidental Tumor Tests	P=0.140	P=0.100N	P=0.174
Hepatocellular Carcinoma			
Overall Rates	17/50 (34%)	9/50 (18%)	21/50 (42%)
Adjusted Rates	40.3%	20.6%	50.2%
Terminal Rates	7/30 (23%)	7/41 (17%)	10/29 (34%)
Life Table Tests	P=0.240	P=0.020N	P=0.298
Incidental Tumor Tests	P=0.160	P=0.112N	P=0.192
Hepatocellular Adenoma or Carcinoma (b)			
Overall Rates	25/50 (50%)	15/50 (30%)	35/50 (70%)
Adjusted Rates	59.5%	33.1%	82.7%
Terminal Rates	14/30 (47%)	11/41 (27%)	22/29 (76%)
Life Table Tests	P=0.044	P=0.007N	P=0.066
Incidental Tumor Tests	P=0.017	P=0.050N	P=0.017
FEMALE			
Hepatocellular Adenoma			
Overall Rates	4/50 (8%)	1/50 (2%)	0/50 (0%)
Adjusted Rates	29.7%	12.5%	0.0%
Terminal Rates	3/12 (25%)	1/8 (13%)	0/9 (0%)
Life Table Tests	P=0.044N	P=0.285N	P=0.092N
Incidental Tumor Tests	P=0.035N	P=0.235N	P=0.072N
Hepatocellular Carcinoma			
Overall Rates	0/50 (0%)	0/50 (0%)	2/50 (4%)
Hepatocellular Adenoma or Carcinoma			
Overall Rates	4/50 (8%)	1/50 (2%)	2/50 (4%)
Adjusted Rates	29.7%	12.5%	14.1%
Terminal Rates	3/12 (25%)	1/8 (13%)	1/9 (11%)
Life Table Tests	P=0.321N	P=0.285N	P=0.429N
Incidental Tumor Tests	P=0.302N	P=0.235N	P=0.396N

(a) The statistical analyses used are discussed in Chapter II (Statistical Methods) and Appendix E (footnotes).
(b) Historical incidence at testing laboratory (mean ± SD): 109/298, 37% ± 12%; historical incidence in NTP studies: 340/1,084, 31% ± 10%

Forestomach: Squamous cell papillomas in female mice occurred with a significant positive trend, although the incidence of squamous cell papillomas in the high dose group was not significantly greater than that in the vehicle controls (Table 18). The occurrence of forestomach epithelial hyperplasia or squamous cell papillomas (combined) was not affected by HC Red No. 3 administration (3/50, 2/50, and 4/48 in vehicle control, low dose, and high dose groups, respectively).

Thyroid Gland: Pigmentation of the thyroid gland was observed at increased incidences in dosed mice of each sex (male: vehicle control, 1/48; low dose, 38/50; high dose, 47/50; female: vehicle control, 2/49; low dose, 13/48; high dose, 24/49). Cystic hyperplasia was observed at increased incidences in dosed male mice (vehicle control, 4/48; low dose, 9/50; high dose, 13/50). The incidence of follicular cell adenomas in low dose male mice was significantly lower ($P < 0.05$) than that in the vehicle controls (Appendix E, Table E3).

Kidney: Nephrosis was observed at increased incidences in dosed female mice (vehicle control, 1/50; low dose, 5/50; high dose, 10/50). These lesions, which involved occasional isolated nephrons in the renal cortex, featured thickened basement membranes, crowding of epithelial cells, cytoplasmic vacuolization, and enlargement and vacuolization of epithelial cell nuclei. Occasionally, portions of the affected tubules were dilated. Early fibrosis and lymphocytic infiltration occurred occasionally in the interstitium of affected areas. The kidneys in the female mice were reviewed in a blind fashion and graded for the presence of nephrosis. The increased incidence of nephrosis was confirmed; however, the apparent effect was ameliorated somewhat by the presence in many mice of glomerulonephritis that appeared to be associated with suppurative inflammation of the genital tract. Thus, the nephrosis may be compound related or secondary to the high incidence of infections. Nephrosis in male mice was comparable in vehicle control and dosed animals.

Multiple Organs: Suppurative inflammation was observed in the uterus, ovary, or multiple organs of 33/38 vehicle control, 37/42 low dose, and 34/41 high dose female mice that died or were killed in a moribund state before the end of the study. Suppurative inflammation of the uterus was observed in 3/12 vehicle control, 0/8 low dose, and 0/9 high dose female mice that lived to the end of the study. Cultures were obtained for one uterine horn and ovary in each of eight female mice that were killed in a moribund condition. Results in six of the eight mice were positive for *Klebsiella pneumoniae*. This infection was believed to be the cause of the reduced survival in female mice.

TABLE 18. ANALYSIS OF FORESTOMACH LESIONS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	Vehicle Control	125 mg/kg	250 mg/kg
Epithelial Hyperplasia			
Overall Rates	3/50 (6%)	2/50 (4%)	1/48 (2%)
Squamous Cell Papilloma (a)			
Overall Rates	0/50 (0%)	0/50 (0%)	3/48 (6%)
Adjusted Rates	0.0%	0.0%	22.8%
Terminal Rates	0/12 (0%)	0/8 (0%)	1/9 (11%)
Life Table Tests	P=0.030	(b)	P=0.092
Incidental Tumor Tests	P=0.031	(b)	P=0.123

(a) Historical incidence of stomach tumors at testing laboratory: 4/297, 1.3%; historical incidence in NTP studies: 7/1,077, 0.6%
(b) No P value is reported because no tumors were observed in the 125 mg/kg and vehicle control groups.

IV. DISCUSSION AND CONCLUSIONS

IV. DISCUSSION AND CONCLUSIONS

Toxicology and carcinogenesis studies of HC Red No. 3, a semipermanent hair dye, were conducted by administering the chemical in corn oil by gavage to groups of 50 male and 50 female F344/N rats for 105 weeks and to groups of 50 male and 50 female $B6C3F_1$ mice for 104 weeks. Groups of 50 rats and 50 mice of each sex served as vehicle controls.

During the planning of the series of studies on hair dyes, the oral route of administration was selected for each chemical in order to provide a more rigorous challenge than would be possible through dermal application. The absorption of HC Red No. 3 through skin has not been studied, but other structurally related dyes (HC Blue No. 1 and HC Blue No. 2) are known to be absorbed in small amounts (C. Burnett, 1984, personal communication to NTP). In the studies of HC Blue No. 1 (NTP, 1985a) and HC Blue No. 2 (NTP, 1985b), the dyes were mixed with the diet; however, HC Red No. 3 was found to be unstable when mixed with feed and therefore was administered by gavage.

The route of administration of these dyes may have influenced the results of the studies. When administered orally, the dyes are exposed to the bacterial flora of the gastrointestinal tract. Nitro reduction, or N-dealkylation, could be carried out by the anaerobic flora of the intestinal tract. These metabolic steps could result in the formation of a free aromatic amine that might be absorbed and then subjected to hepatic N-acetylation and hydroxylation (metabolic steps believed to be associated with the activation of carcinogenic aromatic amines). HC Blue No. 1, which was found to be carcinogenic in rats and mice (NTP, 1985a), was not mutagenic in the in vivo mouse micronucleus test in which chemicals are administered by intraperitoneal injection. This route of administration limits the amount of chemical available for metabolic reduction to a free aromatic amine by the intestinal flora. HC Red No. 3 was not tested in the mouse micronucleus test. HC Blue No. 1, HC Blue No. 2, and HC Red No. 3 are all mutagenic in Salmonella (NTP, 1985a,b; Appendix M).

The doses of HC Red No. 3 administered to rats during the 2-year studies (0, 250, or 500 mg/kg) were selected on the basis of the results of 13-week studies in which rats were dosed with up to 1,000 mg/kg and on the feasibility of gavaging with a viscous suspension. In the 13-week studies, the thyroid gland and kidneys of male and female rats were identified as potential target organs for HC Red No. 3 because of the deposition of a golden-brown pigment (not identified) in the cytoplasm of thyroid gland follicular cells and in the renal convoluted tubular epithelial cells at doses of 250, 500, or 1,000 mg/kg. The intensity of the pigmentation was dose related. Although the accumulated pigment was not considered to be life threatening, there was concern that functional changes might be produced when HC Red No. 3 at 1,000 mg/kg was administered for 2 years. Pigmentation of the kidneys at the 250 mg/kg dose was only slight in both sexes, and there was no pigmentation of the thyroid gland at this dose. Tissues from rats receiving lower doses of HC Red No. 3 were not examined microscopically.

The highest dose of HC Red No. 3 used in both the 13-week and 2-year studies in mice (250 mg/kg) was limited by the viscosity of the corn oil suspension of the dye and the diameter of the gavaging needle. During the 13-week studies, the administration of HC Red No. 3 to mice at doses up to 250 mg/kg did not produce changes in body weight gains or cause clinical signs (other than the excretion of red urine) or histopathologic changes.

The administration of HC Red No. 3 to rats (250 and 500 mg/kg) and mice (125 and 250 mg/kg) for 2 years did not affect body weight gains or survival. In rats, the mean body weights for low dose animals at the termination of the studies were 4% (males) and 5% (females) greater than those of the vehicle control groups. Weight gains by high dose rats were within 10% of the weight gains by the vehicle control groups throughout the studies. In male mice, mean body weights of the low dose group tended to be somewhat lower than those of the vehicle control group; however, mean body weights were always within 10% of the control value, and at the termination of the study, the mean body weight in this group was only 3% lower than that of the vehicle controls. High dose male mice consistently exhibited somewhat greater body weights relative to those of the vehicle controls, and at the termination of the study, the mean body weight of this group was 5% greater than that of

the vehicle control group. The depressed body weight gains exhibited by the low dose female mice are difficult to assess because of the presence of a reproductive tract infection in all groups of females. The body weights of both male and female low dose mice tended to be somewhat lower than those of other groups throughout the study.

The reproductive tract infection in female mice was associated with reduced survival of all groups of female mice; 12/50 vehicle control, 8/50 low dose, and 9/50 high dose animals survived to the end of the study. Uterine horns from eight female mice that were killed in a moribund condition (three vehicle control, two low dose, and three high dose) were examined and subjected to bacteriologic culturing. Results of analyses of six of these tissues were positive for *Klebsiella pneumoniae*. This infection has been present in female $B6C3F_1$ mice in earlier studies at this and other laboratories; both *K. oxytoca* and *K. pneumoniae* have been isolated from tissues of infected animals. The infection appears to be consistently associated with body weight loss and high mortality in NTP studies. The early deaths among female mice may have reduced the sensitivity of the study. The high incidence of early deaths and the absence of a carcinogenic effect render the study in female mice inadequate for assessment of carcinogenicity.

Both rats and mice might have been able to tolerate somewhat higher doses during the studies. Although there was a dose-related increase in the incidence of nephropathy in female rats and nephrosis in female mice, these changes were minimal and, in female mice, were complicated by the presence of genital tract infections.

In mice, there was a dose-related pigmentation of the thyroid glands (male: vehicle control, 1/48; low dose, 38/50; high dose, 47/50; female: vehicle control, 2/49; low dose, 13/48; high dose, 24/49). Although there were no compound-related increases in pathologic changes in the thyroid glands of female mice, there was a dose-related increase in the incidence of cystic hyperplasia in male mice (vehicle control, 4/48; low dose, 9/50; high dose, 13/50). The absence of chemically related toxicologic effects in either species suggests that rats and mice could have tolerated higher doses of HC Red No. 3.

In studies with the semipermanent hair dyes HC Blue No. 1 (NTP, 1985a) and HC Blue No. 2 (1985b), pigmentation of multiple organs, including the thyroid gland and kidneys, was a frequent finding. In the study of HC Blue No. 1, 2-year administration of the dye in the diet was found to produce a dose-related increase in the incidence of cystic hyperplasia of the thyroid gland follicular cells in male mice. However, HC Blue No. 2 did not produce this effect. Neither blue dye produced kidney changes in rats or mice.

Transitional cell papillomas were detected in the urinary bladder of one high dose male rat, two low dose female rats, and one high dose female rat; none was observed in the vehicle controls. The incidences of these tumors in dosed rats and vehicle controls were not significantly different. However, this is an uncommon tumor in F344/N rats, having been detected in 0/299 corn oil vehicle control males and 1/296 (0.3%) corn oil vehicle control females at this laboratory and in 0/1,092 vehicle control males and 3/1,084 (0.3%) vehicle control females in the overall Program (Appendix F, Table F1). The papillomas in the HC Red No. 3 studies were present in animals that survived to the termination of the studies.

The incidence of mammary gland fibroadenomas or cystadenomas (combined) in low dose female rats was significantly increased ($P<0.02$) (vehicle control, 14/50; low dose, 25/50; high dose, 11/50) and was above the historical control rates at the same laboratory (80/300, 27%) and throughout the Program (269/1,147, 23%; Table F2). However, the relationship of this increase to HC Red No. 3 administration is questionable because the incidence in the high dose group was not elevated relative to the vehicle controls and no other chemically related proliferative lesions were noted in the mammary gland. The reason for the increase only in the low dose group is not apparent.

The incidence of hepatocellular adenomas or carcinomas (combined) in male mice was significantly increased in the high dose group (vehicle control, 25/50; low dose, 15/50; high dose, 35/50).

IV. DISCUSSION AND CONCLUSIONS

The arguments for an association of HC Red No. 3 with the hepatocellular neoplasms in male mice include the following:

The incidence in the high dose group was significantly greater than that in the vehicle control group, even though the incidence in the vehicle control group was unusually high (historical rate at the laboratory, 109/298, 37%, or throughout the Program, 340/1,084, 31%).

The incidence of hepatocellular neoplasms in the high dose male mouse group was greater than that seen in any corn oil vehicle control group in the Program.

The increased incidences of hepatocellular neoplasms in dosed male mice were significant by the trend test.

Arguments mitigating the association of HC Red No. 3 with hepatocellular neoplasms in male mice include the following:

Liver neoplasms in control male B6C3F$_1$ mice occur at high incidences and with a wide range of variability (Haseman et al., 1984).

The incidence of hepatocellular neoplasms in low dose male mice was marginally lower than that in the vehicle controls; and the overall rates of liver neoplasms in dosed and vehicle control groups were each 50% (50/100 and 25/50).

The limited data in female mice showed no evidence of a corresponding dose-related increase in the incidence of liver neoplasms.

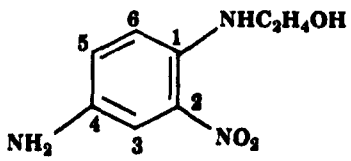
Because of these factors, the increase in hepatocellular neoplasms observed in male mice was considered possibly associated with administration of HC Red No. 3.

HC Red No. 3 is structurally related to both HC Blue No. 1 and HC Blue No. 2, both of which were studied by NTP (Figure 5). Although HC Blue No. 1 produced hepatocellular neoplasms in male and female mice and marginal increases in the incidences of these neoplasms in male rats (NTP, 1985a), no evidence of carcinogenicity was found in NTP studies of HC Blue No. 2 (NTP, 1985b). The difference in the carcinogenicity of the two blue dyes may be due to differences in the way they are metabolized and excreted (NTP, 1985b). The results of the three studies are compared in Table 19.

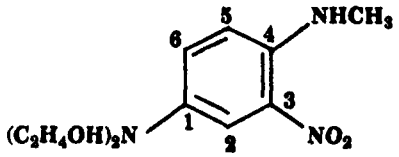
TABLE 19. COMPARISON OF RESULTS IN NTP STUDIES OF HC BLUE NO. 1, HC BLUE NO. 2, AND HC RED NO. 3 IN F344/N RATS AND B6C3F$_1$ MICE

Chemical	Species (sex)	Dose (mg/kg)	Level of Evidence (a)	Organ(s) Affected; Tumor Type
HC Blue No. 1 (Feed)	Rat			
	(male)	(b) 66 or 129	Equivocal	Liver; neoplastic nodules/carcinomas
	(female)	(b) 74 or 154	Some	Lung; alveolar/bronchiolar neoplasms
	Mouse			
	(male)	(b) 309 or 650	Clear	Liver; hepatocellular carcinomas; thyroid gland
	(female)	(b) 778 or 1,634	Clear	Liver; hepatocellular carcinomas
HC Blue No. 2 (Feed)	Rat			
	(male)	(b) 194 or 390	No evidence	
	(female)	(b) 464 or 999	No evidence	
	Mouse			
	(male)	(b) 1,319 or 2,239	No evidence	
	(female)	(b) 2,331 or 5,603	No evidence	
HC Red No. 3 (Gavage)	Rat			
	(male)	250 or 500	No evidence	
	(female)	250 or 500	No evidence	
	Mouse			
	(male)	125 or 250	Equivocal	Liver; adenomas or carcinomas (combined)
	(female)	125 or 250	Inadequate study	

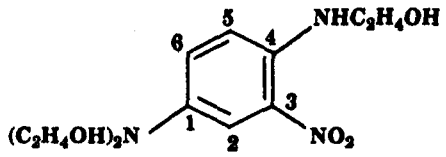
(a) Categories of evidence of carcinogenicity are defined in the Note to the Reader on page 2.
(b) Doses estimated; based on food consumption data.



HC RED NO. 3



HC BLUE NO. 1



HC BLUE NO. 2

FIGURE 5. CHEMICAL STRUCTURES OF HC RED NO. 3, HC BLUE NO. 1, AND HC BLUE NO. 2

IV. DISCUSSION AND CONCLUSIONS

The hydroxyethyl groups on the nitrogens in positions 1 and 4 in HC Blue No. 2 may favor conjugation and urinary excretion, whereas the methyl group on the nitrogen in position 4 of HC Blue No. 1 may favor N-dealkylation and formation of an N-hydroxyl group. In HC Red No. 3, the primary amine in position number 4 may undergo acetylation. However, it is difficult to compare the results of the present studies of HC Red No. 3 with the studies of the blue dyes. In the studies of the blue dyes, the chemical was administered in the diet, whereas the corn oil gavage route of administration was used in the HC Red No. 3 studies. The average daily doses of HC Blue No. 1 administered to high dose rats were 129 and 154 mg/kg in males and females, respectively. HC Red No. 3 was tested at higher doses in rats. In mice, however, the highest doses of HC Blue No. 1 were 650 mg/kg in males and 1,634 mg/kg in females. The highest doses of HC Blue No. 2 tested were 390 and 999 mg/kg in male and female rats and 2,239 and 5,603 mg/kg in male and female mice. Mice received 125 or 250 mg/kg HC Red No. 3 in this study, much less than the amount of either blue dye administered to mice. Also, in the present study, it is possible that each sex of both species could have tolerated higher doses than were administered.

After the HC Red No. 3 studies had ended, the dye samples--lot no. C080480 and lot no. 5890377--used for the 2-year studies were examined for trace contamination with nitrosamines and were found to contain at most 11 and 20 ppm, respectively. Based on maximum possible nitrosamine content of the HC Red No. 3 samples, it was estimated that the high dose rats could have received approximately 10 µg/kg and the high dose mice could have received approximately 5 µg/kg of total nitrosamines per day. Since the nitrosamines were not qualitatively identified, it is impossible to assess their potential impact on the results of these studies. It seems unlikely, however, that these low levels of nitrosamines influenced the results of these studies, since these amounts were equal to (rats) or less than (mice) the amounts of nitrosamines received by dosed animals in the HC Blue No. 2 studies, wherein compound-related increased tumor incidences were not observed. However, this assumes that the nitrosamine contaminants in the HC Red No. 3 study were no more potent as potential carcinogens than were those in the HC Blue No. 2 study.

Conclusions: Under the conditions of these 2-year gavage studies of HC Red No.3, there was *no evidence of carcinogenicity** for male or female F344/N rats given 250 or 500 mg/kg per day. There was *equivocal evidence of carcinogenicity* for male $B6C3F_1$ mice as indicated by an increased incidence of hepatocellular adenomas or carcinomas (combined) in the 250 mg/kg group. Poor survival coupled with lack of significant findings rendered the study in female $B6C3F_1$ mice an *inadequate study of carcinogenicity*. Both sexes of both species may have been able to tolerate higher doses of HC Red No. 3. Therefore, the sensitivity of these studies for detecting carcinogenesis may have been limited.

*Categories of evidence of carcinogenicity are defined in the Note to the Reader on page 2.

V. REFERENCES

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1. Armitage, P. (1971) Statistical Methods in Medical Research. New York: John Wiley & Sons, Inc., pp. 362-365.

2. Berenblum, I., Ed. (1969) Carcinogenicity Testing: A Report of the Panel on Carcinogenicity of the Cancer Research Commission of UICC, Vol. 2. Geneva: International Union Against Cancer.

3. Boorman, G.; Montgomery, C., Jr.; Hardisty, J.; Eustis, S.; Wolfe, M., McConnell, E. (1985) Quality assurance in pathology for rodent toxicology and carcinogenicity tests. Milman, H.; Weisburger, E., Eds.: Handbook of Carcinogen Testing. Park Ridge, NJ: Noyes Publications (in press).

4. Cox, D. (1972) Regression models and life tables. J. R. Stat. Soc. B34:187-220.

5. Feigl, F. (1966) Spot Test in Organic Analysis, 7th ed. Amsterdam: Elsevier Publishing Co., p. 249.

6. Frenkel, E.; Brody, F. (1973) Percutaneous absorption and elimination of an aromatic hair dye. Arch. Environ. Health 27:401-404.

7. Gart, J.; Chu, K.; Tarone, R. (1979) Statistical issues in interpretation of chronic bioassay tests for carcinogenicity. J. Natl. Cancer Inst. 62(4):957-974.

8. Haseman, J. (1984) Statistical issues in the design, analysis and interpretation of animal carcinogenicity studies. Environ. Health Perspect. 58:385-392.

9. Haseman J.; Huff, J.; Boorman, G. (1984) Use of historical control data in carcinogenicity studies in rodents. Toxicol. Path. 12:126-135.

10. Haworth, S.; Lawlor, T.; Mortelmans, K.; Speck, W.; Zeiger, E. (1983) Salmonella mutagenicity test results for 250 chemicals. Environ. Mutagen. (Suppl. 1) 5:3-142.

11. International Agency for Research on Cancer (IARC) (1982) IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans. Some Aromatic Amines, Anthraquinones and Nitroso Compounds, and Inorganic Fluorides Used in Drinking-water and Dental Preparations, Vol. 27. Lyon: World Health Organization, IARC, pp. 307-318.

12. Kaplan, E.; Meier, P. (1958) Nonparametric estimation of incomplete observations. J. Am. Stat. Assoc. 53:457-481.

13. Linhart, M.; Cooper, J.; Martin, R.; Page, N.; Peters, J. (1974) Carcinogenesis bioassay data system. Comp. Biomed. Res. 7:230-248.

14. Mantel, N.; Haenszel, W. (1959) Statistical aspects of the analysis of data from retrospective studies of disease. J. Natl. Cancer Inst. 22:719-748.

15. Maronpot, R.; Boorman, G. (1982) Interpretation of rodent hepatocellular proliferative alterations and hepatocellular tumors in chemical safety assessment. Toxicol. Pathol. 10:71-80.

16. National Cancer Institute (NCI) (1976) Guidelines for Carcinogen Bioassay in Small Rodents. NCI Carcinogenesis Technical Report Series No. 1, February.

17. National Institutes of Health (NIH) (1978) NIH Specification NIH-11-133f, November 1.

18. National Toxicology Program (NTP) (1985a) NTP Technical Report on the Toxicology and Carcinogenesis Studies of HC Blue No. 1 in F344/N Rats and B6C3F$_1$ Mice. NTP TR 271. NIH Publication No. 85-2527. U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health.

19. National Toxicology Program (NTP) (1985b) NTP Technical Report on the Toxicology and Carcinogenesis Studies of HC Blue No. 2 in F344/N Rats and B6C3F$_1$ Mice. NTP TR 293. NIH Publication No. 85-2549. U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health.

20. Tarone, R. (1975) Tests for trend in life table analysis. Biometrika 62:679-682.

21. Wernick, T.; Lanman, B.; Fraux, J. (1975) Chronic toxicity, teratologic, and reproduction studies with hair dyes. Toxicol. Appl. Pharmacol. 32:450-460.

APPENDIX A

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN RATS IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

TABLE A1. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
SQUAMOUS CELL PAPILLOMA		1 (2%)	
SQUAMOUS CELL CARCINOMA	1 (2%)		1 (2%)
BASAL-CELL CARCINOMA		3 (6%)	1 (2%)
SEBACEOUS ADENOCARCINOMA	1 (2%)		1 (2%)
KERATOACANTHOMA	2 (4%)	3 (6%)	2 (4%)
FIBROMA	1 (2%)		
NEURILEMOMA			1 (2%)
*SUBCUT TISSUE	(50)	(50)	(50)
TRICHOEPITHELIOMA		1 (2%)	
FIBROMA	3 (6%)	7 (14%)	2 (4%)
LIPOMA		1 (2%)	
NEUROFIBROSARCOMA	1 (2%)		
RESPIRATORY SYSTEM			
#LUNG	(50)	(50)	(50)
CARCINOMA, NOS, METASTATIC			1 (2%)
SQUAMOUS CELL CARCINOMA	1 (2%)		
ALVEOLAR/BRONCHIOLAR ADENOMA	2 (4%)	1 (2%)	
RHABDOMYOSARCOMA, METASTATIC	1 (2%)		
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
MALIG. LYMPHOMA, LYMPHOCYTIC TYPE	1 (2%)		
LEUKEMIA, MONONUCLEAR CELL	8 (16%)	3 (6%)	3 (6%)
#BONE MARROW	(50)	(50)	(49)
MALIG. LYMPHOMA, HISTIOCYTIC TYPE		1 (2%)	
#LIVER	(50)	(50)	(50)
LEUKEMIA, MONONUCLEAR CELL	1 (2%)		
CIRCULATORY SYSTEM			
*SUBCUT TISSUE	(50)	(50)	(50)
HEMANGIOMA		1 (2%)	1 (2%)
DIGESTIVE SYSTEM			
*SOFT PALATE	(50)	(50)	(50)
SQUAMOUS CELL PAPILLOMA	1 (2%)		1 (2%)
#LIVER	(50)	(50)	(50)
NEOPLASTIC NODULE	3 (6%)		1 (2%)
HEPATOCELLULAR CARCINOMA	1 (2%)	1 (2%)	2 (4%)
#PANCREAS	(50)	(49)	(50)
ACINAR-CELL ADENOMA	11 (22%)	6 (12%)	11 (22%)
ACINAR-CELL CARCINOMA	1 (2%)		
#CECUM	(50)	(50)	(50)
LIPOMA			1 (2%)
URINARY SYSTEM			
#URINARY BLADDER	(50)	(50)	(50)
TRANSITIONAL-CELL PAPILLOMA			1 (2%)

TABLE A1. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM			
#PITUITARY	(50)	(48)	(49)
CARCINOMA, NOS	3 (6%)	2 (4%)	1 (2%)
ADENOMA, NOS	9 (18%)	7 (15%)	7 (14%)
ACIDOPHIL ADENOMA	1 (2%)	2 (4%)	1 (2%)
#ADRENAL	(49)	(50)	(50)
CORTICAL ADENOMA	2 (4%)	1 (2%)	
PHEOCHROMOCYTOMA	19 (39%)	13 (26%)	9 (18%)
PHEOCHROMOCYTOMA, MALIGNANT			1 (2%)
#ADRENAL MEDULLA	(49)	(50)	(50)
PHEOCHROMOCYTOMA	1 (2%)		1 (2%)
GANGLIONEUROMA	1 (2%)		
#THYROID	(49)	(49)	(50)
FOLLICULAR-CELL ADENOMA	1 (2%)	1 (2%)	
FOLLICULAR-CELL CARCINOMA	1 (2%)	2 (4%)	2 (4%)
C-CELL ADENOMA	7 (14%)	5 (10%)	3 (6%)
C-CELL CARCINOMA	5 (10%)		1 (2%)
#PANCREATIC ISLETS	(50)	(49)	(50)
ISLET-CELL ADENOMA	3 (6%)	2 (4%)	3 (6%)
ISLET-CELL CARCINOMA	1 (2%)	1 (2%)	3 (6%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(50)	(50)	(50)
FIBROADENOMA	8 (16%)	2 (4%)	2 (4%)
*PREPUTIAL GLAND	(50)	(50)	(50)
CARCINOMA, NOS		1 (2%)	1 (2%)
ADENOMA, NOS	1 (2%)		1 (2%)
#TESTIS	(50)	(50)	(50)
INTERSTITIAL-CELL TUMOR	46 (92%)	42 (84%)	42 (84%)
*SPERMATIC CORD	(50)	(50)	(50)
MESOTHELIOMA, NOS		1 (2%)	
NERVOUS SYSTEM			
#BRAIN	(50)	(50)	(50)
CARCINOMA, NOS, INVASIVE	1 (2%)	1 (2%)	
SPECIAL SENSE ORGANS			
*ZYMBAL GLAND	(50)	(50)	(50)
CARCINOMA, NOS		1 (2%)	1 (2%)
MUSCULOSKELETAL SYSTEM			
*FEMUR	(50)	(50)	(50)
OSTEOSARCOMA			1 (2%)
*MUSCLE OF THORAX	(50)	(50)	(50)
LIPOMA	1 (2%)		
*ABDOMINAL MUSCLE	(50)	(50)	(50)
LIPOMA		1 (2%)	
*MUSCLE OF LEG	(50)	(50)	(50)
RHABDOMYOSARCOMA	1 (2%)		
BODY CAVITIES			
*PELVIS	(50)	(50)	(50)
LIPOSARCOMA			1 (2%)
*TUNICA VAGINALIS	(50)	(50)	(50)
MESOTHELIOMA, NOS		1 (2%)	1 (2%)

TABLE A1. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(50)	(50)	(50)
SARCOMA, NOS			1 (2%)
MESOTHELIOMA, MALIGNANT			1 (2%)
NEURILEMOMA, MALIGNANT	1 (2%)		
DIAPHRAGM			
SARCOMA, NOS	1		
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	50	50	50
NATURAL DEATH	5	6	8
MORIBUND SACRIFICE	11	9	6
SCHEDULED SACRIFICE			
TERMINAL SACRIFICE	33	33	32
DOSING ACCIDENT	1	2	
ACCIDENTALLY KILLED, NDA			
ACCIDENTALLY KILLED, NOS			4
ANIMAL MISSING			
ANIMAL MISSEXED			
OTHER CASES			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS**	48	48	46
TOTAL PRIMARY TUMORS	152	114	113
TOTAL ANIMALS WITH BENIGN TUMORS	48	47	44
TOTAL BENIGN TUMORS	120	97	89
TOTAL ANIMALS WITH MALIGNANT TUMORS	24	14	17
TOTAL MALIGNANT TUMORS	29	15	22
TOTAL ANIMALS WITH SECONDARY TUMORS##	2	1	1
TOTAL SECONDARY TUMORS	2	1	1
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT	3	1	2
TOTAL UNCERTAIN TUMORS	3	2	2
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* NUMBER OF ANIMALS NECROPSIED
** PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE A2. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
SQUAMOUS CELL PAPILLOMA			1 (2%)
KERATOACANTHOMA	1 (2%)		
*SUBCUT TISSUE	(50)	(50)	(50)
TRICHOEPITHELIOMA	1 (2%)		
SARCOMA, NOS	1 (2%)		
FIBROMA	2 (4%)		2 (4%)
FIBROSARCOMA		1 (2%)	
LIPOMA	1 (2%)		
RESPIRATORY SYSTEM			
#TRACHEA	(50)	(50)	(50)
C-CELL CARCINOMA, INVASIVE			1 (2%)
#LUNG	(50)	(50)	(50)
ADENOCARCINOMA, NOS, METASTATIC		1 (2%)	
ALVEOLAR/BRONCHIOLAR ADENOMA	1 (2%)	1 (2%)	
C-CELL CARCINOMA, METASTATIC			1 (2%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
LEUKEMIA, MONONUCLEAR CELL	10 (20%)	6 (12%)	3 (6%)
#MANDIBULAR L. NODE	(50)	(50)	(50)
SARCOMA, NOS, METASTATIC	1 (2%)		
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(50)	(49)	(50)
SARCOMA, NOS, INVASIVE	1 (2%)		
#PANCREAS	(50)	(50)	(50)
ACINAR-CELL ADENOMA			1 (2%)
#GASTRIC MUCOSA	(50)	(50)	(50)
SQUAMOUS CELL PAPILLOMA		1 (2%)	
#FORESTOMACH	(50)	(50)	(50)
SQUAMOUS CELL PAPILLOMA			2 (4%)
#DUODENUM	(50)	(49)	(49)
LEIOMYOSARCOMA			1 (2%)
URINARY SYSTEM			
#KIDNEY	(50)	(50)	(50)
TUBULAR-CELL ADENOMA	1 (2%)		
#URINARY BLADDER	(50)	(50)	(50)
TRANSITIONAL-CELL PAPILLOMA		2 (4%)	1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(50)	(50)	(50)
CARCINOMA, NOS	4 (8%)	1 (2%)	2 (4%)
ADENOMA, NOS	18 (36%)	17 (34%)	15 (30%)

TABLE A2. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM (Continued)			
#ADRENAL	(50)	(50)	(50)
CORTICAL ADENOMA	2 (4%)	3 (6%)	
CORTICAL CARCINOMA	1 (2%)		
PHEOCHROMOCYTOMA	3 (6%)	3 (6%)	1 (2%)
PHEOCHROMOCYTOMA, MALIGNANT			1 (2%)
#ADRENAL MEDULLA	(50)	(50)	(50)
PHEOCHROMOCYTOMA	1 (2%)		
#THYROID	(50)	(50)	(50)
FOLLICULAR-CELL ADENOMA	1 (2%)	1 (2%)	2 (4%)
FOLLICULAR-CELL CARCINOMA		1 (2%)	1 (2%)
C-CELL ADENOMA	5 (10%)	4 (8%)	4 (8%)
C-CELL CARCINOMA		2 (4%)	2 (4%)
#PARATHYROID	(50)	(48)	(48)
ADENOMA, NOS	1 (2%)		
#PANCREATIC ISLETS	(50)	(50)	(50)
ISLET-CELL ADENOMA		2 (4%)	1 (2%)
ISLET-CELL CARCINOMA		2 (4%)	
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(50)	(50)	(50)
ADENOCARCINOMA, NOS		1 (2%)	2 (4%)
CYSTADENOMA, NOS		1 (2%)	
FIBROADENOMA	14 (28%)	24 (48%)	11 (22%)
*CLITORAL GLAND	(50)	(50)	(50)
CARCINOMA, NOS	1 (2%)	4 (8%)	2 (4%)
ADENOMA, NOS		1 (2%)	
#UTERUS	(50)	(50)	(50)
LEIOMYOMA			1 (2%)
LEIOMYOSARCOMA	1 (2%)		1 (2%)
ENDOMETRIAL STROMAL POLYP	10 (20%)	5 (10%)	9 (18%)
ENDOMETRIAL STROMAL SARCOMA	2 (4%)		
#CERVIX UTERI	(50)	(50)	(50)
FIBROMA		1 (2%)	
ENDOMETRIAL STROMAL POLYP		1 (2%)	
ENDOMETRIAL STROMAL SARCOMA	1 (2%)		
#UTERUS/ENDOMETRIUM	(50)	(50)	(50)
ADENOMA, NOS	1 (2%)		
ADENOCARCINOMA, NOS		1 (2%)	
#OVARY	(50)	(50)	(50)
GRANULOSA-CELL TUMOR		2 (4%)	
NERVOUS SYSTEM			
#BRAIN	(50)	(50)	(50)
CARCINOMA, NOS, INVASIVE			1 (2%)
GRANULAR-CELL TUMOR, NOS		1 (2%)	
SPECIAL SENSE ORGANS			
*ZYMBAL GLAND	(50)	(50)	(50)
CARCINOMA, NOS	1 (2%)		2 (4%)
MUSCULOSKELETAL SYSTEM			
*SKULL	(50)	(50)	(50)
CARCINOMA, NOS, INVASIVE	1 (2%)		

TABLE A2. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
BODY CAVITIES			
*ABDOMINAL WALL	(50)	(50)	(50)
FIBROSARCOMA	1 (2%)		
ALL OTHER SYSTEMS			
NONE			
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	50	50	50
NATURAL DEATH	2	5	11
MORIBUND SACRIFICE	9	6	5
SCHEDULED SACRIFICE			
TERMINAL SACRIFICE	39	38	34
DOSING ACCIDENT			
ACCIDENTALLY KILLED, NDA			
ACCIDENTALLY KILLED, NOS		1	
ANIMAL MISSING			
ANIMAL MISSEXED			
OTHER CASES			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS**	45	44	41
TOTAL PRIMARY TUMORS	86	89	68
TOTAL ANIMALS WITH BENIGN TUMORS	37	40	34
TOTAL BENIGN TUMORS	63	67	51
TOTAL ANIMALS WITH MALIGNANT TUMORS	21	17	14
TOTAL MALIGNANT TUMORS	23	19	17
TOTAL ANIMALS WITH SECONDARY TUMORS##	2	1	2
TOTAL SECONDARY TUMORS	3	1	3
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT		2	
TOTAL UNCERTAIN TUMORS		3	
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* NUMBER OF ANIMALS NECROPSIED
** PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE A3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: VEHICLE CONTROL

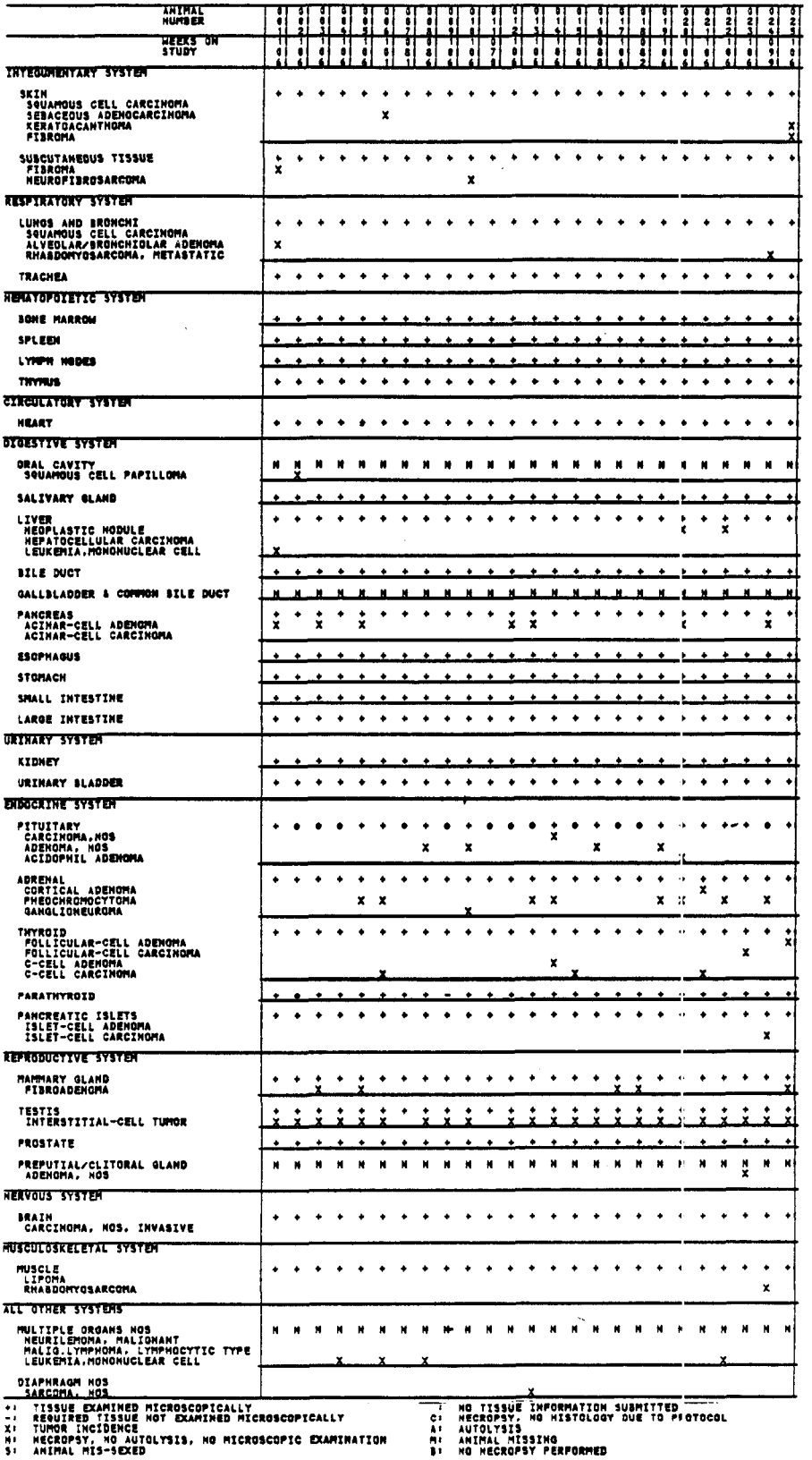

TABLE A3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE RATS: VEHICLE CONTROL (Continued)

ANIMAL NUMBER	0 2 6	0 2 7	0 2 8	0 2 9	0 3 0	0 3 1	0 3 2	0 3 3	0 3 4	0 3 5	0 3 6	0 3 7	0 3 8	0 3 9	0 4 0	0 4 1	0 4 2	0 4 3	0 4 4	0 4 5	0 4 6	0 4 7	0 4 8	0 4 9	0 5 0	TOTAL TISSUES TUMORS
WEEKS ON STUDY	0 6	1 6	1 6	1 6	1 6	1 6	1 6	0 9	0 6	0 6	0 6	0 8	1 6	0 9	0 6	1 6	0 6	0 5	0 5	1 7	1 6	0 6	0 8	0 7	0 6	
INTEGUMENTARY SYSTEM																										
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
SQUAMOUS CELL CARCINOMA							X																			1
SEBACEOUS ADENOCARCINOMA																										1
KERATOACANTHOMA																	X									2
FIBROMA																										1
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
FIBROMA	X																					X				3
NEUROFIBROSARCOMA																										1
RESPIRATORY SYSTEM																										
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SQUAMOUS CELL CARCINOMA																								X		1
ALVEOLAR/BRONCHIOLAR ADENOMA		X																								2
RHABDOMYOSARCOMA, METASTATIC																										1
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEMATOPOIETIC SYSTEM																										
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CIRCULATORY SYSTEM																										
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																										
ORAL CAVITY	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
SQUAMOUS CELL PAPILLOMA																										1
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	49
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
NEOPLASTIC NODULE									X																	3
HEPATOCELLULAR CARCINOMA														X												1
LEUKEMIA, MONONUCLEAR CELL																										1
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ACINAR-CELL ADENOMA	X	X									X				X											11
ACINAR-CELL CARCINOMA	X																									1
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY SYSTEM																										
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ENDOCRINE SYSTEM																										
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA, NOS												X												X		3
ADENOMA, NOS				X		X											X				X	X				9
ACIDOPHIL ADENOMA																										1
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	49
CORTICAL ADENOMA				X																						2
PHEOCHROMOCYTOMA	X	X			X	X			X	X			X	X		X	X					X		X		20
GANGLIONEUROMA																										1
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	49
FOLLICULAR-CELL ADENOMA																										1
FOLLICULAR-CELL CARCINOMA																										1
C-CELL ADENOMA					X	X			X				X		X	X										7
C-CELL CARCINOMA		X					X																			5
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	48
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ISLET-CELL ADENOMA	X			X														X								3
ISLET-CELL CARCINOMA																										1
REPRODUCTIVE SYSTEM																										
MAMMARY GLAND	+	+	+	+	+	+	+	+	M	+	+	+	N	+	+	+	+	+	+	+	+	+	+	+	+	50M
FIBROADENOMA	X													X					X							8
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
INTERSTITIAL-CELL TUMOR	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	46
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
ADENOMA, NOS																										1
NERVOUS SYSTEM																										
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA, NOS, INVASIVE												X														1
MUSCULOSKELETAL SYSTEM																										
MUSCLE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
LIPOMA												X														1
RHABDOMYOSARCOMA																										1
ALL OTHER SYSTEMS																										
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
NEURILEMOMA, MALIGNANT																	X	X								1
MALIG. LYMPHOMA, LYMPHOCYTIC TYPE																X				X	X					1
LEUKEMIA, MONONUCLEAR CELL									X																	8
DIAPHRAGM NOS																										1
SARCOMA, NOS																										

M ANIMALS NECROPSIED

TABLE A3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: LOW DOSE

ANIMAL NUMBER	0 0 1 1	0 0 2 1	0 0 3 1	0 0 4 1	0 0 5 1	0 0 6 1	0 0 7 1	0 0 8 1	0 0 9 1	0 1 0 1	0 1 1 1	0 1 2 1	0 1 3 1	0 1 4 1	0 1 5 1	0 1 6 1	0 1 7 1	0 1 8 1	0 1 9 1	0 2 0 1	0 2 1 1	0 2 2 1	0 2 3 1	0 2 4 1	0 2 5 1
WEEKS ON STUDY	0 8 1	0 8 1	1 0 1	0 8 1	0 9 1	1 0 1	0 8 1	0 8 1	0 8 1	0 9 2	0 8 1	0 8 1	0 6 1	0 9 1	0 8 1	0 8 1	0 8 1	0 8 1	1 0 1	0 8 1	0 9 1	0 8 1	0 9 1	1 0 1	0 7 1
INTEGUMENTARY SYSTEM																									
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+
SQUAMOUS CELL PAPILLOMA																									
BASAL-CELL CARCINOMA															X				X	X					
KERATOACANTHOMA																									
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+
TRICHOEPITHELIOMA																							X		
FIBROMA										X							X	X							
LIPOMA																									
HEMANGIOMA																									
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ALVEOLAR/BRONCHIOLAR ADENOMA									X																
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																									
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEPATOCELLULAR CARCINOMA																						X			
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ACINAR-CELL ADENOMA															X			X							
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CARCINOMA,NOS	X												X												
ADENOMA, NOS																					X				
ACIDOPHIL ADENOMA																								X	
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CORTICAL ADENOMA								X		X												X		X	
PHEOCHROMOCYTOMA																									
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
FOLLICULAR-CELL ADENOMA						X																			
FOLLICULAR-CELL CARCINOMA												X					X								
C-CELL ADENOMA	X																								
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ISLET-CELL ADENOMA		X															X								
ISLET-CELL CARCINOMA															X										
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	+	+	+	N	+	+	+	+	+	N
FIBROADENOMA								X																	
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
INTERSTITIAL-CELL TUMOR		X	X	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X			X	X		
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
CARCINOMA,NOS																		X							
VAS DEFERNES, SPERMATIC CORD	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
MESOTHELIOMA, NOS																									
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CARCINOMA, NOS, INVASIVE													X												
SPECIAL SENSE ORGANS																									
ZYMBAL'S GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	+	M	M	M	M	M	M	M	M	M	M	M
CARCINOMA,NOS																									
MUSCULOSKELETAL SYSTEM																									
MUSCLE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LIPOMA															X										
BODY CAVITIES																									
TUNICA VAGINALIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
MESOTHELIOMA, NOS																									
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
LEUKEMIA,MONONUCLEAR CELL			X										X												

TABLE A3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE RATS: LOW DOSE (Continued)

ANIMAL NUMBER / WEEKS ON STUDY	TOTAL TISSUES TUMORS
INTEGUMENTARY SYSTEM	
SKIN	50M
SQUAMOUS CELL PAPILLOMA	1
BASAL-CELL CARCINOMA	3
KERATOACANTHOMA	3
SUBCUTANEOUS TISSUE	50M
TRICHOEPITHELIOMA	1
FIBROMA	7
LIPOMA	1
HEMANGIOMA	1
RESPIRATORY SYSTEM	
LUNGS AND BRONCHI	50
ALVEOLAR/BRONCHIOLAR ADENOMA	1
TRACHEA	50
HEMATOPOIETIC SYSTEM	
BONE MARROW	50
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	1
SPLEEN	49
LYMPH NODES	50
THYMUS	50
CIRCULATORY SYSTEM	
HEART	50
DIGESTIVE SYSTEM	
SALIVARY GLAND	50
LIVER	50
HEPATOCELLULAR CARCINOMA	1
BILE DUCT	50
GALLBLADDER & COMMON BILE DUCT	50M
PANCREAS	49
ACINAR-CELL ADENOMA	6
ESOPHAGUS	50
STOMACH	50
SMALL INTESTINE	49
LARGE INTESTINE	50
URINARY SYSTEM	
KIDNEY	50
URINARY BLADDER	50
ENDOCRINE SYSTEM	
PITUITARY	48
CARCINOMA,NOS	2
ADENOMA, NOS	7
ACIDOPHIL ADENOMA	2
ADRENAL	50
CORTICAL ADENOMA	1
PHEOCHROMOCYTOMA	13
THYROID	49
FOLLICULAR-CELL ADENOMA	1
FOLLICULAR-CELL CARCINOMA	2
C-CELL ADENOMA	5
PARATHYROID	49
PANCREATIC ISLETS	49
ISLET-CELL ADENOMA	2
ISLET-CELL CARCINOMA	1
REPRODUCTIVE SYSTEM	
MAMMARY GLAND	50M
FIBROADENOMA	2
TESTIS	50
INTERSTITIAL-CELL TUMOR	42
PROSTATE	50
PREPUTIAL/CLITORAL GLAND	50M
CARCINOMA,NOS	1
VAS DEFERNES, SPERMATIC CORD	50M
MESOTHELIOMA, NOS	1
NERVOUS SYSTEM	
BRAIN	50
CARCINOMA, NOS, INVASIVE	1
SPECIAL SENSE ORGANS	
ZYMBAL'S GLAND	50M
CARCINOMA,NOS	1
MUSCULOSKELETAL SYSTEM	
MUSCLE	50M
LIPOMA	1
BODY CAVITIES	
TUNICA VAGINALIS	50M
MESOTHELIOMA, NOS	1
ALL OTHER SYSTEMS	
MULTIPLE ORGANS NOS	50M
LEUKEMIA,MONONUCLEAR CELL	1

* ANIMALS NECROPSIED

TABLE A3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: HIGH DOSE

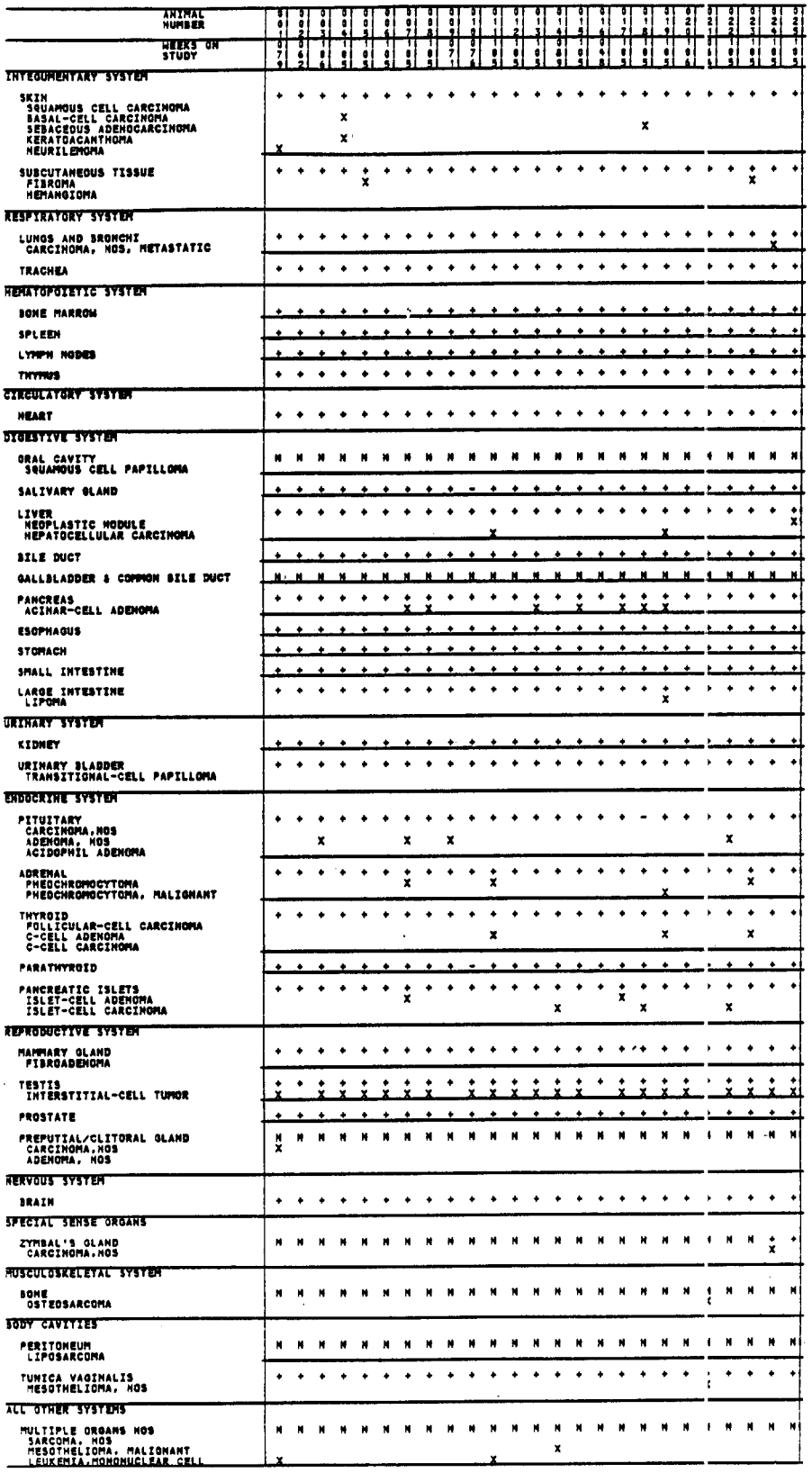

TABLE A3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE RATS: HIGH DOSE (Continued)

ANIMAL NUMBER	0 2 4 6	0 2 7 6	0 2 8 6	0 3 2 6	0 3 3 6	0 3 3 7	0 3 3 7	0 3 3 7	0 3 4 6	0 3 5 6	0 3 5 6	0 3 5 6	0 3 5 6	0 3 5 6	0 3 5 6	0 4 1 1	0 4 1 2	0 4 1 2	0 4 1 3	0 4 1 5	0 4 1 5	0 4 1 7	0 4 1 8	0 4 1 9	0 4 1 9	0 5 0 0	TOTAL TISSUES TUMORS
WEEKS ON STUDY	0 4 1	0 4 7	0 4 1	0 4 1	0 4 5	0 5 5	0 4 1	0 4 1	0 7 5	0 7 9	0 8 1	0 8 1	0 8 1	0 8 1	0 8 1	0 8 1	0 8 1	0 9 2	0 9 3	0 9 3	1 0 4	1 0 4	0 9 4	0 9 4	1 0 5	0 8 7	
INTEGUMENTARY SYSTEM																											
SKIN	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	X	+	+	+	+	+	50M
SQUAMOUS CELL CARCINOMA																											1
BASAL-CELL CARCINOMA																											1
SEBACEOUS ADENOCARCINOMA																											1
KERATOACANTHOMA																									X		2
NEURILEMOMA																											1
SUBCUTANEOUS TISSUE	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
FIBROMA				X																							2
HEMANGIOMA																											1
RESPIRATORY SYSTEM																											
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA, NOS, METASTATIC																											1
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEMATOPOIETIC SYSTEM																											
BONE MARROW	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
THYMUS	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
CIRCULATORY SYSTEM																											
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																											
ORAL CAVITY	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
SQUAMOUS CELL PAPILLOMA													X														1
SALIVARY GLAND	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
NEOPLASTIC NODULE																											1
HEPATOCELLULAR CARCINOMA																											2
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ACINAR-CELL ADENOMA	X			X			X						X														11
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	49
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LIPOMA																											1
URINARY SYSTEM																											
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TRANSITIONAL-CELL PAPILLOMA					X																						1
ENDOCRINE SYSTEM																											
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
CARCINOMA,NOS				X																							1
ADENOMA, NOS															X										X	X	7
ACIDOPHIL ADENOMA					X																						1
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
PHEOCHROMOCYTOMA	X							X							X			X	X	X		X					10
PHEOCHROMOCYTOMA, MALIGNANT																											1
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FOLLICULAR-CELL CARCINOMA																	X							X			2
C-CELL ADENOMA							X																				3
C-CELL CARCINOMA																											1
PARATHYROID	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	46
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	X	+	+	+	50
ISLET-CELL ADENOMA																											1
ISLET-CELL CARCINOMA																											3
REPRODUCTIVE SYSTEM																											
MAMMARY GLAND	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	X	+	+	+	50M
FIBROADENOMA	X																										2
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
INTERSTITIAL-CELL TUMOR	X	X	X	X	X		X	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X		X	42
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
CARCINOMA,NOS		X																									1
ADENOMA, NOS																											1
NERVOUS SYSTEM																											
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPECIAL SENSE ORGANS																											
ZYMBAL'S GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
CARCINOMA,NOS																											1
MUSCULOSKELETAL SYSTEM																											
BONE	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
OSTEOSARCOMA																											1
BODY CAVITIES																											
PERITONEUM	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
LIPOSARCOMA									X																		1
TUNICA VAGINALIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
MESOTHELIOMA, NOS																											1
ALL OTHER SYSTEMS																											
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
SARCOMA, NOS																										X	1
MESOTHELIOMA, MALIGNANT																											1
LEUKEMIA,MONONUCLEAR CELL																			X								1

M ANIMALS NECROPSIED

TABLE A4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: VEHICLE CONTROL

ANIMAL NUMBER	0 0 0 1	0 0 2 1	0 0 3 1	0 0 4 1	0 0 5 1	0 0 6 1	0 0 7 1	0 0 8 1	0 0 9 1	0 1 0 1	0 1 1 1	0 1 2 1	0 1 3 1	0 1 4 1	0 1 5 1	0 1 6 1	0 1 7 1	0 1 8 1	0 1 9 1	0 2 0 1	0 2 1 1	0 2 2 1	0 2 3 1	0 2 4 1	0 2 5 1
WEEKS ON STUDY	0 4 1	0 7 1	1 0 1	1 0 1	1 2 1	1 3 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1	1 4 1
INTEGUMENTARY SYSTEM																									
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
KERATOACANTHOMA																									
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	X	+	+	+	+	+	+	+	+	+	+
TRICHOEPITHELIOMA																									
SARCOMA, NOS						X																			
FIBROMA																						X			
LIPOMA																									
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ALVEOLAR/BRONCHIOLAR ADENOMA																									
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SARCOMA, NOS, METASTATIC						X																			
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SARCOMA, NOS, INVASIVE						X																			
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
TUBULAR-CELL ADENOMA																									
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CARCINOMA,NOS													X												X
ADENOMA, NOS		X	X				X		X	X	X	X			X				X				X		
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CORTICAL ADENOMA		X																							
CORTICAL CARCINOMA																									
PHEOCHROMOCYTOMA								X	X																
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FOLLICULAR-CELL ADENOMA																	X								
C-CELL ADENOMA	X																	X							
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOMA, NOS																						X			
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FIBROADENOMA		X		X											X	X	X	X		X				X	
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
CARCINOMA,NOS																									
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	X	+	+	+	+	+
ADENOMA, NOS																									
LEIOMYOSARCOMA																									
ENDOMETRIAL STROMAL POLYP	X					X									X		X		X						
ENDOMETRIAL STROMAL SARCOMA					X	X																X			
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+						
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPECIAL SENSE ORGANS																									
ZYMBAL'S GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	+
CARCINOMA,NOS																									X
MUSCULOSKELETAL SYSTEM																									
BONE	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
CARCINOMA, NOS, INVASIVE																									
BODY CAVITIES																									
PERITONEUM	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
FIBROSARCOMA																									
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
LEUKEMIA,MONONUCLEAR CELL											X		X				X					X			

+: TISSUE EXAMINED MICROSCOPICALLY
-: REQUIRED TISSUE NOT EXAMINED MICROSCOPICALLY
X: TUMOR INCIDENCE
M: NECROPSY, NO AUTOLYSIS, NO MICROSCOPIC EXAMINATION
S: ANIMAL MIS-SEXED
I: NO TISSUE INFORMATION SUBMITTED
C: NECROPSY, NO HISTOLOGY DUE TO PROTOCOL
A: AUTOLYSIS
M: ANIMAL MISSING
B: NO NECROPSY PERFORMED

TABLE A4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE RATS: VEHICLE CONTROL

(Continued)

ANIMAL NUMBER	0 2 6 0	0 3 7 1	0 3 8 2	0 4 0 3	0 4 5 4	0 4 5 5	0 4 5 6	0 4 5 7	0 4 5 8	0 4 5 9	0 4 6 0	0 4 6 1	0 4 6 2	0 4 6 3	0 4 6 4	0 4 6 5	0 4 6 6	0 4 6 7	0 4 6 8	0 4 6 9	0 4 7 0	TOTAL TISSUES TUMORS
WEEKS ON STUDY	0 8	0 8	0 8	0 9	0 8	0 8	0 8	0 8	0 8	0 8	0 8	0 8	0 9	0 8	0 8	0 8	0 8	0 8	0 8	0 8	0 8	
INTEGUMENTARY SYSTEM																						
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
KERATOACANTHOMA													X									1
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TRICHOEPITHELIOMA																						1
SARCOMA, NOS																						1
FIBROMA									X											X		2
LIPOMA																						1
RESPIRATORY SYSTEM																						
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ALVEOLAR/BRONCHIOLAR ADENOMA																			X			1
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEMATOPOIETIC SYSTEM																						
BONE MARROW	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SARCOMA, NOS, METASTATIC																						1
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CIRCULATORY SYSTEM																						
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																						
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SARCOMA, NOS, INVASIVE																						1
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY SYSTEM																						
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TUBULAR-CELL ADENOMA																					X	1
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ENDOCRINE SYSTEM																						
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA, NOS																	X					4
ADENOMA, NOS	X		X		X			X			X	X	X						X	X	X	18
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CORTICAL ADENOMA						X																2
CORTICAL CARCINOMA				X										X								1
PHEOCHROMOCYTOMA																	X					4
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FOLLICULAR-CELL ADENOMA					X													X		X		1
C-CELL ADENOMA																			X			5
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOMA, NOS																						1
REPRODUCTIVE SYSTEM																						
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FIBROADENOMA			X								X			X				X	X	X		14
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
CARCINOMA, NOS																					X	1
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOMA, NOS																						1
LEIOMYOSARCOMA			X	X			X					X			X							10
ENDOMETRIAL STROMAL POLYP														X			X					3
ENDOMETRIAL STROMAL SARCOMA																						1
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
NERVOUS SYSTEM																						
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPECIAL SENSE ORGANS																						
ZYMBAL'S GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	+	M	M	M	M	50M
CARCINOMA, NOS																						1
MUSCULOSKELETAL SYSTEM																						
BONE	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
CARCINOMA, NOS, INVASIVE																	X					1
BODY CAVITIES																						
PERITONEUM	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
FIBROSARCOMA												X										1
ALL OTHER SYSTEMS																						
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
LEUKEMIA, MONONUCLEAR CELL			X	X			X						X		X					X		10

M ANIMALS NECROPSIED

TABLE A4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: LOW DOSE

ANIMAL NUMBER	0 0 1	0 0 2	0 0 3	0 0 4	0 0 5	0 0 6	0 0 7	0 0 8	0 0 9	0 1 0	0 1 1	0 1 2	0 1 3	0 1 4	0 1 5	0 1 6	0 1 7	0 1 8	0 1 9	0 2 0	0 2 1	0 2 2	0 2 3	0 2 4	0 2 5
WEEKS ON STUDY	0 6 5	0 8 6	0 8 6	0 8 6	0 8 6	0 9 1	0 8 6	0 8 5	0 8 6	0 8 6	0 8 6	0 8 6	0 8 6	0 7 8	0 8 9	0 8 2	0 8 6	0 9 2	0 8 6	0 8 6	0 8 6	0 6 0	0 9 4	0 8 6	0 8 6
INTEGUMENTARY SYSTEM																									
SUBCUTANEOUS TISSUE FIBROSARCOMA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI ADENOCARCINOMA, NOS, METASTATIC	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ALVEOLAR/BRONCHIOLAR ADENOMA	X																				X				
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SQUAMOUS CELL PAPILLOMA																									
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
TRANSITIONAL-CELL PAPILLOMA		X																							
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CARCINOMA, NOS															X										
ADENOMA, NOS					X	X	X		X			X	X	X									X	X	X
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CORTICAL ADENOMA																				X					
PHEOCHROMOCYTOMA											X					X									
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FOLLICULAR-CELL ADENOMA										X															
FOLLICULAR-CELL CARCINOMA																									
C-CELL ADENOMA																							X		
C-CELL CARCINOMA																									X
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	+
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ISLET-CELL ADENOMA			X																						
ISLET-CELL CARCINOMA										X															
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOCARCINOMA, NOS	X																								
CYSTADENOMA, NOS																							X		
FIBROADENOMA			X	X	X				X		X				X	X	X				X			X	
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	X	X	X	M	M	X	M	M	M	M
CARCINOMA, NOS																X	X	X			X				
ADENOMA, NOS																									
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOCARCINOMA, NOS																									
FIBROMA			X																						
ENDOMETRIAL STROMAL POLYP			X																		X				
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GRANULOSA-CELL TUMOR												X													
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GRANULAR-CELL TUMOR, NOS																									
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
LEUKEMIA, MONONUCLEAR CELL																	X					X			

TABLE A4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE RATS: LOW DOSE (Continued)

ANIMAL NUMBER	026	027	028	029	031	032	033	034	035	036	037	038	039	040	041	042	043	044	045	046	047	048	049	050	TOTAL TISSUES TUMORS
WEEKS ON STUDY	106	106	106	118	098	106	106	106	106	106	106	106	086	086	106	099	106	106	106	106	106	106	106	106	
INTEGUMENTARY SYSTEM																									
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
FIBROSARCOMA																X									1
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOCARCINOMA, NOS, METASTATIC																									1
ALVEOLAR/BRONCHIOLAR ADENOMA																									1
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SQUAMOUS CELL PAPILLOMA																			X						1
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TRANSITIONAL-CELL PAPILLOMA													X												2
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA,NOS																									1
ADENOMA, NOS	X	X			X						X				X								X	X	17
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CORTICAL ADENOMA															X								X		3
PHEOCHROMOCYTOMA																							X		1
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FOLLICULAR-CELL ADENOMA																			X						1
FOLLICULAR-CELL CARCINOMA																			X						1
C-CELL ADENOMA																	X	X		X					4
C-CELL CARCINOMA				X																					2
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ISLET-CELL ADENOMA	X				X																				2
ISLET-CELL CARCINOMA																									2
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
ADENOCARCINOMA, NOS																									1
CYSTADENOMA, NOS																									1
FIBROADENOMA	X	X		X	X	X				X	X		X	X			X	X			X		X	X	24
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
CARCINOMA,NOS						X																			4
ADENOMA, NOS																									1
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOCARCINOMA, NOS										X															1
FIBROMA																									1
ENDOMETRIAL STROMAL POLYP		X				X	X																	X	6
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GRANULOSA-CELL TUMOR																								X	2
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GRANULAR-CELL TUMOR, NOS																								X	1
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
LEUKEMIA,MONONUCLEAR CELL				X					X													X			4

M ANIMALS NECROPSIED

TABLE A4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: HIGH DOSE

ANIMAL NUMBER	001	002	003	004	005	006	007	008	009	010	011	012	013	014	015	016	017	018	019	020	021	022	023	024	025
WEEKS ON STUDY	106	106	106	092	106	106	106	106	080	106	106	106	106	106	106	106	106	112	095	096	095	095	106	095	095
INTEGUMENTARY SYSTEM																									
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SQUAMOUS CELL PAPILLOMA																									
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FIBROMA													X												
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-CELL CARCINOMA, METASTATIC										X															
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-CELL CARCINOMA, INVASIVE										X															
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ACINAR-CELL ADENOMA																									
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SQUAMOUS CELL PAPILLOMA																								X	
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+
LEIOMYOSARCOMA																									
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
TRANSITIONAL-CELL PAPILLOMA																				X					
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CARCINOMA,NOS																									
ADENOMA, NOS	X	X	X							X										X			X	X	
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
PHEOCHROMOCYTOMA																+									
PHEOCHROMOCYTOMA, MALIGNANT																X									
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FOLLICULAR-CELL ADENOMA																							X		
FOLLICULAR-CELL CARCINOMA																									
C-CELL ADENOMA		X	X						X							X				X					
C-CELL CARCINOMA										X															
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ISLET-CELL ADENOMA									X																
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOCARCINOMA, NOS										X															
FIBROADENOMA					X							X	X				X			X			X		
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
CARCINOMA,NOS																									
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LEIOMYOMA																				X					
LEIOMYOSARCOMA																									
ENDOMETRIAL STROMAL POLYP								X			X		X					X		X					
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
CARCINOMA, NOS, INVASIVE																									
SPECIAL SENSE ORGANS																									
ZYMBAL'S GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	+	M	M	M	M	M	M
CARCINOMA,NOS																			X						
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M
LEUKEMIA,MONONUCLEAR CELL																		X					X		

TABLE A4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE RATS: HIGH DOSE (Continued)

ANIMAL NUMBER	0226	0227	0228	0229	0230	0231	0232	0233	0234	0235	0236	0237	0238	0239	0240	0241	0242	0243	0244	0245	0246	0247	0248	0249	0250	TOTAL TISSUES TUMORS
WEEKS ON STUDY	078	086	096	096	096	096	096	079	096	096	096	086	086	091	096	096	096	096	086	096	096	084	097	096		
INTEGUMENTARY SYSTEM																										
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	+	+	50M
SQUAMOUS CELL PAPILLOMA											X															1
SUBCUTANEOUS TISSUE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	+	+	50M
FIBROMA																						X				2
RESPIRATORY SYSTEM																										
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
C-CELL CARCINOMA, METASTATIC																										1
TRACHEA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
C-CELL CARCINOMA, INVASIVE																										1
HEMATOPOIETIC SYSTEM																										
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
THYMUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CIRCULATORY SYSTEM																										
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																										
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ACINAR-CELL ADENOMA			X																							1
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SQUAMOUS CELL PAPILLOMA																								X		2
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
LEIOMYOSARCOMA								X																		1
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	49
URINARY SYSTEM																										
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TRANSITIONAL-CELL PAPILLOMA																										1
ENDOCRINE SYSTEM																										
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA,NOS							X													X						2
ADENOMA, NOS				X		X				X		X		X				X			X			X		15
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
PHEOCHROMOCYTOMA																										1
PHEOCHROMOCYTOMA, MALIGNANT																								X		1
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FOLLICULAR-CELL ADENOMA	X			X																						2
FOLLICULAR-CELL CARCINOMA																										1
C-CELL ADENOMA																										4
C-CELL CARCINOMA																										2
PARATHYROID	-	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	46
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ISLET-CELL ADENOMA																										1
REPRODUCTIVE SYSTEM																										
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
ADENOCARCINOMA, NOS													X		X		X	X		X						2
FIBROADENOMA																								X		11
PREPUTIAL/CLITORAL GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
CARCINOMA,NOS			X																						X	2
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LEIOMYOMA																						X				1
LEIOMYOSARCOMA		X			X								X					X								1
ENDOMETRIAL STROMAL POLYP																										9
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
NERVOUS SYSTEM																										
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
CARCINOMA, NOS, INVASIVE																				X						1
SPECIAL SENSE ORGANS																										
ZYMBAL'S GLAND	+	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	50M
CARCINOMA,NOS	X																									2
ALL OTHER SYSTEMS																										
MULTIPLE ORGANS NOS	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	50M
LEUKEMIA,MONONUCLEAR CELL									X																	3

M ANIMALS NECROPSIED

APPENDIX B

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

TABLE B1. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
FIBROMA		2 (4%)	1 (2%)
NEUROFIBROMA		1 (2%)	
RESPIRATORY SYSTEM			
#LUNG	(50)	(49)	(50)
HEPATOCELLULAR CARCINOMA, METAST	3 (6%)	1 (2%)	5 (10%)
ALVEOLAR/BRONCHIOLAR ADENOMA	6 (12%)	7 (14%)	7 (14%)
ALVEOLAR/BRONCHIOLAR CARCINOMA	6 (12%)	7 (14%)	7 (14%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
MALIG. LYMPHOMA, LYMPHOCYTIC TYPE		2 (4%)	1 (2%)
MALIG. LYMPHOMA, HISTIOCYTIC TYPE	1 (2%)	1 (2%)	1 (2%)
MALIGNANT LYMPHOMA, MIXED TYPE	6 (12%)	5 (10%)	3 (6%)
LYMPHOCYTIC LEUKEMIA		1 (2%)	
#BRONCHIAL LYMPH NODE	(50)	(49)	(50)
ALVEOLAR/BRONCHIOLAR CA, METASTA			1 (2%)
#MESENTERIC L. NODE	(50)	(49)	(50)
MALIG. LYMPHOMA, HISTIOCYTIC TYPE		1 (2%)	1 (2%)
#PEYER'S PATCH	(50)	(48)	(50)
MALIG. LYMPHOMA, LYMPHOCYTIC TYPE		1 (2%)	
CIRCULATORY SYSTEM			
#SPLEEN	(50)	(50)	(50)
HEMANGIOSARCOMA	4 (8%)		2 (4%)
#LIVER	(50)	(50)	(50)
HEMANGIOSARCOMA	4 (8%)	2 (4%)	2 (4%)
DIGESTIVE SYSTEM			
#SUBMAXILLARY GLAND	(50)	(50)	(50)
SQUAMOUS CELL CARCINOMA	1 (2%)		
#LIVER	(50)	(50)	(50)
HEPATOCELLULAR ADENOMA	11 (22%)	6 (12%)	16 (32%)
HEPATOCELLULAR CARCINOMA	17 (34%)	9 (18%)	21 (42%)
CARCINOID TUMOR, METASTATIC			1 (2%)
*COMMON BILE DUCT	(50)	(50)	(50)
CARCINOID TUMOR, MALIGNANT			1 (2%)
#FORESTOMACH	(50)	(50)	(50)
SQUAMOUS CELL PAPILLOMA			1 (2%)
#JEJUNUM	(50)	(48)	(50)
ADENOCARCINOMA, NOS	1 (2%)		1 (2%)
URINARY SYSTEM			
#KIDNEY	(50)	(50)	(50)
TUBULAR-CELL ADENOCARCINOMA	1 (2%)		
#URINARY BLADDER	(49)	(49)	(50)
TRANSITIONAL-CELL PAPILLOMA			1 (2%)

TABLE B1. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM			
#ADRENAL	(50)	(50)	(50)
PHEOCHROMOCYTOMA	1 (2%)		
#ADRENAL/CAPSULE	(50)	(50)	(50)
ADENOMA, NOS	2 (4%)	4 (8%)	3 (6%)
#THYROID	(48)	(50)	(50)
FOLLICULAR-CELL ADENOMA	8 (17%)	3 (6%)	5 (10%)
#PANCREATIC ISLETS	(50)	(50)	(50)
ISLET-CELL ADENOMA	1 (2%)		4 (8%)
REPRODUCTIVE SYSTEM			
*SEMINAL VESICLE	(50)	(50)	(50)
ALVEOLAR/BRONCHIOLAR CA, METASTA			1 (2%)
#TESTIS	(50)	(50)	(50)
INTERSTITIAL-CELL TUMOR		2 (4%)	
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
*HARDERIAN GLAND	(50)	(50)	(50)
ADENOMA, NOS	2 (4%)		3 (6%)
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*THORAX	(50)	(50)	(50)
NEUROFIBROSARCOMA		1 (2%)	
*MEDIASTINUM	(50)	(50)	(50)
ALVEOLAR/BRONCHIOLAR CA, METASTA			1 (2%)
*MESENTERY	(50)	(50)	(50)
SARCOMA, NOS			1 (2%)
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(50)	(50)	(50)
SARCOMA, NOS, METASTATIC			1 (2%)
NEUROFIBROSARCOMA		1 (2%)	
LUMBAR REGION			
NEUROFIBROSARCOMA		1	

TABLE B1. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	50	50	50
NATURAL DEATH	8	4	3
MORIBUND SACRIFICE	12	5	18
SCHEDULED SACRIFICE			
TERMINAL SACRIFICE	30	41	29
DOSING ACCIDENT			
ACCIDENTALLY KILLED, NDA			
ACCIDENTALLY KILLED, NOS			
ANIMAL MISSING			
ANIMAL MISSEXED			
OTHER CASES			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS**	44	36	42
TOTAL PRIMARY TUMORS	72	57	82
TOTAL ANIMALS WITH BENIGN TUMORS	22	17	31
TOTAL BENIGN TUMORS	31	25	41
TOTAL ANIMALS WITH MALIGNANT TUMORS	32	27	30
TOTAL MALIGNANT TUMORS	41	32	41
TOTAL ANIMALS WITH SECONDARY TUMORS##	3	1	9
TOTAL SECONDARY TUMORS	3	1	10
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT			
TOTAL UNCERTAIN TUMORS			
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* NUMBER OF ANIMALS NECROPSIED
** PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE B2. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SUBCUT TISSUE	(50)	(50)	(50)
SEBACEOUS ADENOMA			1 (2%)
RESPIRATORY SYSTEM			
#LUNG	(50)	(49)	(50)
ALVEOLAR/BRONCHIOLAR ADENOMA		1 (2%)	1 (2%)
ALVEOLAR/BRONCHIOLAR CARCINOMA	1 (2%)	1 (2%)	
OSTEOSARCOMA, METASTATIC	1 (2%)		
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
MALIG. LYMPHOMA, LYMPHOCYTIC TYPE	1 (2%)	2 (4%)	
MALIGNANT LYMPHOMA, MIXED TYPE	3 (6%)	1 (2%)	6 (12%)
#PANCREATIC L. NODE	(50)	(49)	(50)
MALIGNANT LYMPHOMA, MIXED TYPE		1 (2%)	
#LIVER	(50)	(50)	(50)
MALIG. LYMPHOMA, HISTIOCYTIC TYPE		1 (2%)	
CIRCULATORY SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
HEMANGIOSARCOMA		1 (2%)	
*PELVIC ORGANS	(50)	(50)	(50)
HEMANGIOSARCOMA			1 (2%)
#LIVER	(50)	(50)	(50)
HEMANGIOSARCOMA	1 (2%)		
DIGESTIVE SYSTEM			
#LIVER	(50)	(50)	(50)
HEPATOCELLULAR ADENOMA	4 (8%)	1 (2%)	
HEPATOCELLULAR CARCINOMA			2 (4%)
#FORESTOMACH	(50)	(50)	(48)
SQUAMOUS CELL PAPILLOMA			3 (6%)
#JEJUNUM	(48)	(49)	(50)
ADENOMA, NOS	1 (2%)		
ADENOCARCINOMA, NOS	1 (2%)		
URINARY SYSTEM			
NONE			
ENDOCRINE SYSTEM			
#PITUITARY	(47)	(45)	(43)
ADENOMA, NOS	4 (9%)	2 (4%)	6 (14%)
#ADRENAL	(50)	(49)	(49)
PHEOCHROMOCYTOMA	1 (2%)		
OSTEOSARCOMA, METASTATIC	1 (2%)		
#ADRENAL/CAPSULE	(50)	(49)	(49)
ADENOMA, NOS		1 (2%)	
#ADRENAL MEDULLA	(50)	(49)	(49)
PHEOCHROMOCYTOMA		1 (2%)	

TABLE B2. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM (Continued)			
#THYROID	(49)	(48)	(49)
FOLLICULAR-CELL ADENOMA	2 (4%)	1 (2%)	2 (4%)
FOLLICULAR-CELL CARCINOMA			1 (2%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(50)	(50)	(50)
ADENOMA, NOS			1 (2%)
ADENOCARCINOMA, NOS		2 (4%)	
#UTERUS	(50)	(50)	(50)
LEIOMYOMA	1 (2%)		
ENDOMETRIAL STROMAL POLYP	2 (4%)	1 (2%)	
#OVARY	(45)	(48)	(48)
GRANULOSA-CELL TUMOR	1 (2%)		
TERATOMA, NOS			1 (2%)
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
*HARDERIAN GLAND	(50)	(50)	(50)
ADENOMA, NOS			1 (2%)
MUSCULOSKELETAL SYSTEM			
*RIB	(50)	(50)	(50)
OSTEOSARCOMA	1 (2%)		
BODY CAVITIES			
*MEDIASTINUM	(50)	(50)	(50)
ALVEOLAR/BRONCHIOLAR CA, INVASIVE		1 (2%)	
ALL OTHER ORGANS			
*MULTIPLE ORGANS	(50)	(50)	(50)
SARCOMA, NOS	1 (2%)		1 (2%)
FIBROSARCOMA	1 (2%)		
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	50	50	50
NATURAL DEATH	24	22	19
MORIBUND SACRIFICE	14	20	22
SCHEDULED SACRIFICE			
TERMINAL SACRIFICE	12	8	9
DOSING ACCIDENT			
ACCIDENTALLY KILLED, NDA			
ACCIDENTALLY KILLED, NOS			
ANIMAL MISSING			
ANIMAL MISSEXED			
OTHER CASES			

TABLE B2. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS**	16	15	18
TOTAL PRIMARY TUMORS	26	17	27
TOTAL ANIMALS WITH BENIGN TUMORS	12	7	10
TOTAL BENIGN TUMORS	15	8	15
TOTAL ANIMALS WITH MALIGNANT TUMORS	9	8	11
TOTAL MALIGNANT TUMORS	10	9	11
TOTAL ANIMALS WITH SECONDARY TUMORS##	1	1	
TOTAL SECONDARY TUMORS	2	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			

• NUMBER OF ANIMALS NECROPSIED
** PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE B3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: VEHICLE CONTROL

ANIMAL NUMBER	0011	0012	0013	0014	0015	0016	0017	0018	0019	0020	0111	0112	0113	0114	0115	0116	0117	0118	0119	0120	0121	0122	0123	0124	0125
WEEKS ON STUDY	105	105	105	105	091	105	105	076	086	090	105	105	086	075	081	105	105	105	094	091	105	094	106	095	105
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEPATOCELLULAR CARCINOMA, METASTA					X				X							X									
ALVEOLAR/BRONCHIOLAR ADENOMA			X	X		X									X										
ALVEOLAR/BRONCHIOLAR CARCINOMA		X	X				X					X					X							X	
TRACHEA	-	-	+	-	+	-	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEMANGIOSARCOMA								X																X	
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
THYMUS	+	+	+	-	-	+	+	+	+	+	+	-	+	+	-	+	-	+	+	+	-	+	+	+	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SQUAMOUS CELL CARCINOMA																						X			
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEPATOCELLULAR ADENOMA			X		X		X						X				X								
HEPATOCELLULAR CARCINOMA			X		X				X		X			X	X					X			X		
HEMANGIOSARCOMA								X											X		X				X
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	M	+	+	+	M	+	+	+	+	+	+	+	+	+	M	M	+	+	M	+	+	+	+	+	+
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOCARCINOMA, NOS																									
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
TUBULAR-CELL ADENOCARCINOMA																									
URINARY BLADDER	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOMA, NOS																									
PHEOCHROMOCYTOMA																									
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+
FOLLICULAR-CELL ADENOMA				X		X											X	X							
PARATHYROID	-	-	+	-	+	-	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ISLET-CELL ADENOMA																									
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPECIAL SENSE ORGANS																									
HARDERIAN GLAND	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
ADENOMA, NOS	X																								
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
MALIG.LYMPHOMA, HISTIOCYTIC TYPE											X														
MALIGNANT LYMPHOMA, MIXED TYPE													X												X

+: TISSUE EXAMINED MICROSCOPICALLY
-: REQUIRED TISSUE NOT EXAMINED MICROSCOPICALLY
X: TUMOR INCIDENCE
N: NECROPSY, NO AUTOLYSIS, NO MICROSCOPIC EXAMINATION
S: ANIMAL MIS-SEXED
I: NO TISSUE INFORMATION SUBMITTED
C: NECROPSY, NO HISTOLOGY DUE TO PROTOCOL
A: AUTOLYSIS
M: ANIMAL MISSING
B: NO NECROPSY PERFORMED

TABLE B3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE MICE: VEHICLE CONTROL (Continued)

ANIMAL NUMBER	0 2 6	0 2 7	0 2 8	0 2 9	0 3 1	0 3 2	0 3 3	0 3 4	0 3 5	0 3 6	0 3 7	0 3 8	0 3 9	0 4 0	0 4 1	0 4 2	0 4 3	0 4 4	0 4 5	0 4 6	0 4 7	0 4 8	0 4 9	0 5 0	TOTAL TISSUES TUMORS
WEEKS ON STUDY	1 1 0	0 5	0 4 5	0 5	1 0 5	1 2 5	1 3 5	1 4 5	1 5 5	1 6 5	1 8 2	1 8 5	1 9 4	1 9 5	1 9 5	1 9 5	1 9 5	1 9 5	1 9 5	1 9 5	1 9 5	1 9 5	1 9 5	1 9 6	
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEPATOCELLULAR CARCINOMA, METASTA											X	X													3
ALVEOLAR/BRONCHIOLAR ADENOMA																									6
ALVEOLAR/BRONCHIOLAR CARCINOMA																									4
TRACHEA	+	-	-	+	+	+	-	-	-	-	+	+	-	-	+	+	-	-	+	+	-	+	+	-	-	31
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEMANGIOSARCOMA							X										X								4
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
THYMUS	+	+	+	-	+	+	-	+	-	-	-	+	+	+	+	+	-	+	+	+	-	+	-	+	35
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SQUAMOUS CELL CARCINOMA																									1
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEPATOCELLULAR ADENOMA		X				X	X		X					X		X		X							11
HEPATOCELLULAR CARCINOMA									X	X						X					X	X	X		17
HEMANGIOSARCOMA																X									4
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	M	+	+	+	+	M	+	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	49
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOCARCINOMA, NOS																						X			1
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TUBULAR-CELL ADENOCARCINOMA														X											1
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
ENDOCRINE SYSTEM																									
PITUITARY	+	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	46
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOMA, NOS	X				X																				2
PHEOCHROMOCYTOMA														X											1
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	48
FOLLICULAR-CELL ADENOMA	X						X							X		X									2
PARATHYROID	+	-	-	+	-	+	-	-	-	-	+	+	+	-	+	+	+	-	+	-	-	+	-	-	-	30
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ISLET-CELL ADENOMA														X											1
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPECIAL SENSE ORGANS																									
HARDERIAN GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
ADENOMA, NOS				X																					2
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																								X	1
MALIGNANT LYMPHOMA, MIXED TYPE		X										X							X						4

M ANIMALS NECROPSIED

TABLE B3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: LOW DOSE

ANIMAL NUMBER	0 0 0 0 0 0 0 0 0 0 0 0 1 1 1 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2 2 2 2 2
WEEKS ON STUDY	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
INTEGUMENTARY SYSTEM	
SKIN	+ + + + N +
FIBROMA	X
NEUROFIBROMA	
RESPIRATORY SYSTEM	
LUNGS AND BRONCHI	+ + + + + - +
HEPATOCELLULAR CARCINOMA, METASTA	X
ALVEOLAR/BRONCHIOLAR ADENOMA	X X X
ALVEOLAR/BRONCHIOLAR CARCINOMA	X
TRACHEA	- - - - - + - - - - - - - - + - + - - + - - + + - -
HEMATOPOIETIC SYSTEM	
BONE MARROW	+ +
SPLEEN	+ +
LYMPH NODES	+ +
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	
THYMUS	- + + + + - + + - - - + + - + - + + + + + + + + -
CIRCULATORY SYSTEM	
HEART	+ +
DIGESTIVE SYSTEM	
SALIVARY GLAND	+ +
LIVER	+ +
HEPATOCELLULAR ADENOMA	X X X X X X
HEPATOCELLULAR CARCINOMA	X
HEMANGIOSARCOMA	
BILE DUCT	+ +
GALLBLADDER & COMMON BILE DUCT	+ + + N + N N + N + + + + + + + N + + + + + + N + + + + + + + + + + + +
PANCREAS	+ +
ESOPHAGUS	+ +
STOMACH	+ +
SMALL INTESTINE	- +
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE	X
LARGE INTESTINE	+ +
URINARY SYSTEM	
KIDNEY	+ +
URINARY BLADDER	+ + + + + - +
ENDOCRINE SYSTEM	
PITUITARY	+ + + + + + + - +
ADRENAL	+ +
ADENOMA, NOS	X X X
THYROID	+ +
FOLLICULAR-CELL ADENOMA	X X
PARATHYROID	- - - - + + - - + - - + + + - + + + + - - + + -
REPRODUCTIVE SYSTEM	
MAMMARY GLAND	N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N
TESTIS	+ +
INTERSTITIAL-CELL TUMOR	X
PROSTATE	+ +
NERVOUS SYSTEM	
BRAIN	+ +
BODY CAVITIES	
PLEURA	N N N N N N N N N N N N N N N N N I N N N N N N N N N N N N N N N N N N
NEUROFIBROSARCOMA	
ALL OTHER SYSTEMS	
MULTIPLE ORGANS NOS	N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N N
NEUROFIBROSARCOMA	
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE	X
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	X
MALIGNANT LYMPHOMA, MIXED TYPE	X X
LYMPHOCYTIC LEUKEMIA	
LUMBAR REGION	
NEUROFIBROSARCOMA	

TABLE B3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE MICE: LOW DOSE (Continued)

ANIMAL NUMBER	0226	0227	0231	0251	0256	0311	0312	0334	0335	0336	0337	0338	0339	0340	0341	0342	0343	0344	0345	0346	0347	0348	0349	0350	TOTAL TISSUES TUMORS
WEEKS ON STUDY	0 5	0 5	0 5	0 5	0 5	0 5	0 6	0 7	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 5	0 4	
INTEGUMENTARY SYSTEM																									
SKIN	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
FIBROMA																X									2
NEUROFIBROMA																									1
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
HEPATOCELLULAR CARCINOMA, METASTA					X																				1
ALVEOLAR/BRONCHIOLAR ADENOMA				X		X			X							X	X	X							7
ALVEOLAR/BRONCHIOLAR CARCINOMA									X				X										X		7
TRACHEA	-	-	-	-	-	-	+	-	-	-	-	-	-	+	+	-	+	-	+	+	+	-	+	+	16
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	49
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																							X		1
THYMUS	+	+	+	-	+	+	+	-	-	+	+	-	+	+	-	+	+	+	+	+	+	-	-	+	36
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	49
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LIVER	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEPATOCELLULAR ADENOMA			X						X							X			X		X				6
HEPATOCELLULAR CARCINOMA				X	X					X			X									X	X		9
HEMANGIOSARCOMA																									2
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	+	+	+	+	M	M	+	+	+	+	M	+	M	+	+	+	+	+	+	+	+	+	+	M	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE																									1
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	47
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOMA, NOS						X														X					4
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FOLLICULAR-CELL ADENOMA																X									3
PARATHYROID	+	-	-	+	+	-	-	+	+	+	+	+	+	+	+	-	+	-	+	+	+	-	+	+	31
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
INTERSTITIAL-CELL TUMOR				X																					2
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
BODY CAVITIES																									
PLEURA	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
NEUROFIBROSARCOMA																			X						1
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
NEUROFIBROSARCOMA							X																		1
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE								X																	2
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																									1
MALIGNANT LYMPHOMA, MIXED TYPE		X	X	X																					5
LYMPHOCYTIC LEUKEMIA																									1
LUMBAR REGION																									
NEUROFIBROSARCOMA									X																1

M ANIMALS NECROPSIED

TABLE B3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: HIGH DOSE

ANIMAL NUMBER	001	002	003	004	005	006	007	008	009	010	011	012	013	014	015	016	017	018	019	020	021	022	023	024	025
WEEKS ON STUDY	088	105	105	092	098	091	085	109	085	099	085	085	085	085	085	048	095	095	087	093	085	085	095	095	095
INTEGUMENTARY SYSTEM																									
SKIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	N	+	+	+	+	+	+	+	+	+
FIBROMA																								X	
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEPATOCELLULAR CARCINOMA, METASTA	X															X									
ALVEOLAR/BRONCHIOLAR ADENOMA					X	X				X															
ALVEOLAR/BRONCHIOLAR CARCINOMA																X									
TRACHEA	+	+	-	+	-	+	-	-	+	+	-	+	-	+	-	+	-	-	-	-	+	-	-	-	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEMANGIOSARCOMA										X										X					
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ALVEOLAR/BRONCHIOLAR CA, METASTAT													X												
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																									
THYMUS	+	+	-	-	-	+	-	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	-	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEPATOCELLULAR ADENOMA	X		X	X		X	X			X	X		X	X		X	X	X	X	X			X	X	
HEPATOCELLULAR CARCINOMA																									X
CARCINOID TUMOR, METASTATIC																					X				
HEMANGIOSARCOMA																									
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+
CARCINOID TUMOR, MALIGNANT																									
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SQUAMOUS CELL PAPILLOMA																									
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOCARCINOMA, NOS																									X
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
TRANSITIONAL-CELL PAPILLOMA																			X						
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	-	+	+	-	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOMA, NOS																			X						
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FOLLICULAR-CELL ADENOMA				X																		X		X	
PARATHYROID	+	+	+	-	-	+	+	-	-	+	-	+	+	+	+	-	+	+	-	-	+	-	-	-	-
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ISLET-CELL ADENOMA										X										X					
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SEMINAL VESICLE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ALVEOLAR/BRONCHIOLAR CA, METASTAT					X																				
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPECIAL SENSE ORGANS																									
HARDERIAN GLAND	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
ADENOMA, NOS	X																X					X			
BODY CAVITIES																									
MEDIASTINUM	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
ALVEOLAR/BRONCHIOLAR CA, METASTAT																									
MESENTERY	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
SARCOMA, NOS																									
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
SARCOMA, NOS, METASTATIC														X											
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE																									
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																									
MALIGNANT LYMPHOMA, MIXED TYPE															X					X					X

TABLE B3. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF MALE MICE: HIGH DOSE (Continued)

ANIMAL NUMBER	0261	0271	0281	0291	0301	0311	0321	0331	0341	0351	0361	0371	0381	0391	0401	0411	0421	0431	0441	0451	0461	0471	0481	0491	0501	TOTAL TISSUES TUMORS
WEEKS ON STUDY	077	091	105	103	105	105	105	105	096	091	090	105	085	111	101	101	086	093	105	105	096	096	105	105	105	
INTEGUMENTARY SYSTEM																										
SKIN	+	+	+	+	+	N	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
FIBROMA																										1
RESPIRATORY SYSTEM																										
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEPATOCELLULAR CARCINOMA, METASTA										X	X		X													5
ALVEOLAR/BRONCHIOLAR ADENOMA	X	X							X		X				X					X					X	7
ALVEOLAR/BRONCHIOLAR CARCINOMA			X																	X	X					7
TRACHEA	+	+	+	+	+	-	-	+	+	+	+	-	-	+	-	-	-	+	-	+	+	+	-	-	+	26
HEMATOPOIETIC SYSTEM																										
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEMANGIOSARCOMA																										2
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ALVEOLAR/BRONCHIOLAR CA, METASTAT																					X					1
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																										1
THYMUS	+	-	+	-	+	+	+	+	-	-	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	39
CIRCULATORY SYSTEM																										
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																										
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEPATOCELLULAR ADENOMA			X		X			X							X		X		X			X		X	16
HEPATOCELLULAR CARCINOMA	X								X	X	X	X	X			X		X						X		21
CARCINOID TUMOR, METASTATIC		X																								1
HEMANGIOSARCOMA																					X					2
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	N	+	+	+	+	N	+	+	+	N	50M
CARCINOID TUMOR, MALIGNANT		X																								1
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SQUAMOUS CELL PAPILLOMA			X																							1
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOCARCINOMA, NOS																										1
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY SYSTEM																										
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
TRANSITIONAL-CELL PAPILLOMA																										1
ENDOCRINE SYSTEM																										
PITUITARY	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	-	+	+	+	+	44
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ADENOMA, NOS												X													X	3
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
FOLLICULAR-CELL ADENOMA						X				X																5
PARATHYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+	36
PANCREATIC ISLETS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ISLET-CELL ADENOMA																					X		X			4
REPRODUCTIVE SYSTEM																										
MAMMARY GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
TESTIS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
PROSTATE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SEMINAL VESICLE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50M
ALVEOLAR/BRONCHIOLAR CA, METASTAT																										1
NERVOUS SYSTEM																										
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPECIAL SENSE ORGANS																										
HARDERIAN GLAND	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
ADENOMA, NOS																										3
BODY CAVITIES																										
MEDIASTINUM	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
ALVEOLAR/BRONCHIOLAR CA, METASTAT																					X					1
MESENTERY	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
SARCOMA, NOS	X																									1
ALL OTHER SYSTEMS																										
MULTIPLE ORGANS NOS	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	M	50M
SARCOMA, NOS, METASTATIC	X																									1
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE																		X								1
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																										1
MALIGNANT LYMPHOMA, MIXED TYPE																										1

M ANIMALS NECROPSIED

TABLE B4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: VEHICLE CONTROL

ANIMAL NUMBER	0 0 0 0 0 0 0 0 0 0 0 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2 2 2
WEEKS ON STUDY	0 0 0 0 0 0 0 0 0 1 1 0 0 0 0 0 0 0 0 0 0 0 0 0 1 1 1 1 1 1
	8 8 9 7 9 7 9 7 7 0 0 0 0 0 0 0 0 0 0 0 5 7 6 9 5 7 7 8
RESPIRATORY SYSTEM	
LUNGS AND BRONCHI	+ +
ALVEOLAR/BRONCHIOLAR CARCINOMA	X X
OSTEOSARCOMA, METASTATIC	X
TRACHEA	- + + + + - - + + + + + + - - + + - - - + + - - + +
HEMATOPOIETIC SYSTEM	
BONE MARROW	+ +
SPLEEN	+ +
LYMPH NODES	+ +
THYMUS	+ + + + + + + - + - + + + + - + + + + - + + + + - -
CIRCULATORY SYSTEM	
HEART	+ +
DIGESTIVE SYSTEM	
SALIVARY GLAND	+ +
LIVER	+ +
HEPATOCELLULAR ADENOMA	X
HEMANGIOSARCOMA	X
BILE DUCT	+ +
GALLBLADDER & COMMON BILE DUCT	+ + N + + + N + + + + + + + + N N + N + N + + + + + N
PANCREAS	+ +
ESOPHAGUS	+ +
STOMACH	+ +
SMALL INTESTINE	+ +
ADENOMA, NOS	X
ADENOCARCINOMA, NOS	X
LARGE INTESTINE	+ - +
URINARY SYSTEM	
KIDNEY	+ +
URINARY BLADDER	+ +
ENDOCRINE SYSTEM	
PITUITARY	+ +
ADENOMA, NOS	X X X X X
ADRENAL	+ +
PHEOCHROMOCYTOMA	X
OSTEOSARCOMA, METASTATIC	X
THYROID	+ +
FOLLICULAR-CELL ADENOMA	X
PARATHYROID	- + + + - - + + + + + + + + + + - - - - + + - + +
REPRODUCTIVE SYSTEM	
MAMMARY GLAND	+ + + + + + + + N + N + + + + + + + + + + + + + + + +
UTERUS	+ +
LEIOMYOMA	X
ENDOMETRIAL STROMAL POLYP	X
OVARY	+ + + + + + - + + + - + + + + + + + + + + + + + + + +
GRANULOSA-CELL TUMOR	X
NERVOUS SYSTEM	
BRAIN	+ +
MUSCULOSKELETAL SYSTEM	
BONE	M M M M M M M M M M M M M M M M M M M M M M M M M M M M
OSTEOSARCOMA	X
ALL OTHER SYSTEMS	
MULTIPLE ORGANS NOS	M M M M M M M M M M M M M M M M M M M M M M M M M M M M
SARCOMA, NOS	X
FIBROSARCOMA	X
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE	X X X
MALIGNANT LYMPHOMA, MIXED TYPE	X

+: TISSUE EXAMINED MICROSCOPICALLY
-: REQUIRED TISSUE NOT EXAMINED MICROSCOPICALLY
X: TUMOR INCIDENCE
N: NECROPSY, NO AUTOLYSIS, NO MICROSCOPIC EXAMINATION
S: ANIMAL MIS-SEXED
: NO TISSUE INFORMATION SUBMITTED
C: NECROPSY, NO HISTOLOGY DUE TO PROTOCOL
A: AUTOLYSIS
M: ANIMAL MISSING
B: NO NECROPSY PERFORMED

TABLE B4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE MICE: VEHICLE CONTROL

(Continued)

ANIMAL NUMBER / WEEKS ON STUDY	TOTAL TISSUES / TUMORS
RESPIRATORY SYSTEM	
LUNGS AND BRONCHI	50
ALVEOLAR/BRONCHIOLAR CARCINOMA	1
OSTEOSARCOMA, METASTATIC	1
TRACHEA	29
HEMATOPOIETIC SYSTEM	
BONE MARROW	50
SPLEEN	50
LYMPH NODES	50
THYMUS	41
CIRCULATORY SYSTEM	
HEART	50
DIGESTIVE SYSTEM	
SALIVARY GLAND	49
LIVER	50
HEPATOCELLULAR ADENOMA	4
HEMANGIOSARCOMA	1
BILE DUCT	50
GALLBLADDER & COMMON BILE DUCT	50M
PANCREAS	50
ESOPHAGUS	50
STOMACH	50
SMALL INTESTINE	48
ADENOMA, NOS	1
ADENOCARCINOMA, NOS	1
LARGE INTESTINE	49
URINARY SYSTEM	
KIDNEY	50
URINARY BLADDER	50
ENDOCRINE SYSTEM	
PITUITARY	47
ADENOMA, NOS	4
ADRENAL	50
PHEOCHROMOCYTOMA	1
OSTEOSARCOMA, METASTATIC	1
THYROID	49
FOLLICULAR-CELL ADENOMA	2
PARATHYROID	32
REPRODUCTIVE SYSTEM	
MAMMARY GLAND	50M
UTERUS	50
LEIOMYOMA	1
ENDOMETRIAL STROMAL POLYP	2
OVARY	45
GRANULOSA-CELL TUMOR	1
NERVOUS SYSTEM	
BRAIN	50
MUSCULOSKELETAL SYSTEM	
BONE	50M
OSTEOSARCOMA	1
ALL OTHER SYSTEMS	
MULTIPLE ORGANS NOS	50M
SARCOMA, NOS	1
FIBROSARCOMA	1
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE	1
MALIGNANT LYMPHOMA, MIXED TYPE	3

M ANIMALS NECROPSIED

TABLE B4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: LOW DOSE

ANIMAL NUMBER	0011	0012	0013	0014	0015	0016	0017	0018	0019	0020	0021	0022	0023	0024	0025	0026	0027	0028	0029	0030	0031	0032	0033	0034	0035
WEEKS ON STUDY	0935	0887	0861	0879	0869	0887	0977	0985	0991	0991	0971	0995	0881	0874	0885	0883	0870	0857	0881	0880	0894	0890	0915	0899	0877
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ALVEOLAR/BRONCHIOLAR ADENOMA							X																		
TRACHEA	+	+	+	-	-	+	+	-	+	+	+	+	-	+	+	-	+	-	-	-	+	-	+	+	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
MALIGNANT LYMPHOMA, MIXED TYPE																									
THYMUS	+	+	+	+	+	-	+	+	+	+	+	-	-	+	-	-	+	+	+	+	+	-	+	+	+
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
HEPATOCELLULAR ADENOMA																							X		
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																									
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	+	+	+	+	N	+	+	+	+	+	+	+	+	N	+	+	N	+	+	+	+	N	+	+	+
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+
ADENOMA, NOS	X																								
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOMA, NOS																							X		
PHEOCHROMOCYTOMA																									
THYROID	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
FOLLICULAR-CELL ADENOMA																			X						
PARATHYROID	+	+	-	-	-	+	+	-	+	+	+	+	-	+	+	+	+	-	-	-	+	-	+	+	+
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ADENOCARCINOMA, NOS													X											X	
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ENDOMETRIAL STROMAL POLYP																									
OVARY	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
BODY CAVITIES																									
MEDIASTINUM	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
ALVEOLAR/BRONCHIOLAR CA, INVASIVE							X																		
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
HEMANGIOSARCOMA													X												
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE										X															
MALIGNANT LYMPHOMA, MIXED TYPE																									

TABLE B4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE MICE: LOW DOSE (Continued)

ANIMAL NUMBER	0226	0227	0228	0229	0301	0302	0303	0304	0305	0306	0307	0308	0309	0310	0401	0402	0403	0404	0405	0406	0407	0408	0409	0410	TOTAL TISSUES TUMORS
WEEKS ON STUDY	086	078	081	081	064	069	071	078	081	091	081	086	086	087	085	074	064	071	063	048	084	087	081	085	
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	49
ALVEOLAR/BRONCHIOLAR ADENOMA																X									1
ALVEOLAR/BRONCHIOLAR CARCINOMA																									1
TRACHEA	-	+	-	+	-	-	-	+	-	-	+	-	+	-	+	-	-	+	-	+	+	+	-	+	27
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	49
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	49
MALIGNANT LYMPHOMA, MIXED TYPE														X											1
THYMUS	+	+	+	+	+	+	+	+	+	-	+	+	+	+	-	+	+	-	+	+	+	+	+	-	46
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LIVER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
HEPATOCELLULAR ADENOMA															X										1
MALIG.LYMPHOMA, HISTIOCYTIC TYPE																									1
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	N	+	+	+	+	+	+	+	+	50N
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	49
STOMACH	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SMALL INTESTINE	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	49
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ENDOCRINE SYSTEM																									
PITUITARY	+	+	+	+	+	-	+	-	+	+	+	+	+	+	-	+	+	+	+	-	+	+	+	+	45
ADENOMA, NOS				X																					2
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	49
ADENOMA, NOS										X															1
PHEOCHROMOCYTOMA																									1
THYROID	+	+	+	+	+	+	-	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	48
FOLLICULAR-CELL ADENOMA																									1
PARATHYROID	+	+	-	+	-	-	-	+	-	-	+	+	+	-	-	+	+	+	-	-	+	+	+	-	38
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	+	50N
ADENOCARCINOMA, NOS																									2
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	X	+	+	+	+	+	+	+	50
ENDOMETRIAL STROMAL POLYP																									1
OVARY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	48
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
BODY CAVITIES																									
MEDIASTINUM	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	50N
ALVEOLAR/BRONCHIOLAR CA, INVASIVE																									1
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	50N
HEMANGIOSARCOMA																							X		1
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE																								X	2
MALIGNANT LYMPHOMA, MIXED TYPE																									1

N ANIMALS NECROPSIED

TABLE B4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3: HIGH DOSE

ANIMAL NUMBER	0 0 1	0 0 2	0 0 3	0 0 4	0 0 5	0 0 6	0 0 7	0 0 8	0 0 9	0 1 0	0 1 1	0 1 2	0 1 3	0 1 4	0 1 5	0 1 6	0 1 7	0 1 8	0 1 9	0 2 0	0 2 1	0 2 2	0 2 3	0 2 4	0 2 5
WEEKS ON STUDY	0 5 9	0 7 6	0 7 6	1 0 2	0 6 7	0 7 2	0 5 7	1 0 5	0 9 5	0 8 6	0 7 4	0 8 6	0 8 7	0 8 4	0 4 9	0 6 8	0 7 0	0 6 0	0 5 3	0 8 0	1 0 5	0 9 4	1 0 5	0 7 4	0 6 0
INTEGUMENTARY SYSTEM																									
SUBCUTANEOUS TISSUE SEBACEOUS ADENOMA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	+	+
RESPIRATORY SYSTEM																									
LUNGS AND BRONCHI ALVEOLAR/BRONCHIOLAR ADENOMA	+	+	+	+	+	+	+	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
TRACHEA	-	+	-	+	-	-	-	-	-	-	-	-	-	+	+	+	-	+	+	-	-	-	+	-	-	+
HEMATOPOIETIC SYSTEM																									
BONE MARROW	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
THYMUS	+	+	+	+	+	-	+	+	+	+	+	+	-	+	+	-	-	-	+	+	+	+	+	+	-
CIRCULATORY SYSTEM																									
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
DIGESTIVE SYSTEM																									
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+
LIVER HEPATOCELLULAR CARCINOMA	+	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
GALLBLADDER & COMMON BILE DUCT	N	+	+	+	+	+	+	+	+	+	+	N	+	+	+	+	+	+	+	+	+	+	+	N	+
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STOMACH SQUAMOUS CELL PAPILLOMA	+	+	+	+	+	+	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY SYSTEM																									
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
URINARY BLADDER	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
ENDOCRINE SYSTEM																									
PITUITARY ADENOMA, NOS	+	+	-	+ X	+	-	+	+	+ X	+	-	-	+ X	+	+	+	+	+	-	+	+	+	+	+	+
ADRENAL	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
THYROID FOLLICULAR-CELL ADENOMA FOLLICULAR-CELL CARCINOMA	+	+	+	+	+	+	+	+ X	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
PARATHYROID	-	+	-	+	-	-	+	-	-	-	-	-	+	+	-	-	+	+	-	-	+	+	+	-	+
REPRODUCTIVE SYSTEM																									
MAMMARY GLAND ADENOMA, NOS	N	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	N	+	+	N	+	+	+	+
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
OVARY TERATOMA, NOS	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
NERVOUS SYSTEM																									
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SPECIAL SENSE ORGANS																									
HARDERIAN GLAND ADENOMA, NOS	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N
BODY CAVITIES																									
PERITONEUM HEMANGIOSARCOMA	N	N	N	N	N	N	N	N	N	N	N X	N	N	N	N	N	N	N	N	N	N	N	N	N	N
ALL OTHER SYSTEMS																									
MULTIPLE ORGANS NOS SARCOMA, NOS MALIGNANT LYMPHOMA, MIXED TYPE	N	N X	N	N	N	N	N	N X	N	N	N	N	N	N	N	N	N	N	N	N	N X	N	N X	N	N

TABLE B4. INDIVIDUAL ANIMAL TUMOR PATHOLOGY OF FEMALE MICE: HIGH DOSE (Continued)

ANIMAL NUMBER	026	027	028	029	030	031	032	033	034	035	036	037	038	039	040	041	042	043	044	045	046	047	048	049	050	TOTAL TISSUES TUMORS
WEEKS ON STUDY	079	084	105	105	130	151	068	059	075	076	088	094	105	105	098	090	086	081	050	057	069	094	096	081	070	
INTEGUMENTARY SYSTEM																										
SUBCUTANEOUS TISSUE SEBACEOUS ADENOMA	+	+	+	+	N	+	+	+	+	N	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	50N 1
RESPIRATORY SYSTEM																										
LUNGS AND BRONCHI ALVEOLAR/BRONCHIOLAR ADENOMA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50 1
TRACHEA	-	-	+	-	+	-	+	+	-	-	+	+	-	-	+	+	-	+	-	-	-	-	+	-	+	20
HEMATOPOIETIC SYSTEM																										
BONE MARROW	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
SPLEEN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LYMPH NODES	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
THYMUS	+	+	-	+	-	+	+	-	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	-	39
CIRCULATORY SYSTEM																										
HEART	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
DIGESTIVE SYSTEM																										
SALIVARY GLAND	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
LIVER HEPATOCELLULAR CARCINOMA	+	+	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50 2
BILE DUCT	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
GALLBLADDER & COMMON BILE DUCT	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	M	+	50M
PANCREAS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
ESOPHAGUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
STOMACH SQUAMOUS CELL PAPILLOMA	+	+	+	+	+	+	+	+	+	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+ X	-	+	48 1
SMALL INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
LARGE INTESTINE	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
URINARY SYSTEM																										
KIDNEY	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
URINARY BLADDER	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
ENDOCRINE SYSTEM																										
PITUITARY ADENOMA, NOS	-	+ X	+	+	+	+	+	+	+	-	+	+ X	+ X	+	+	+	+	+	+	+	+	+	+	+	+	43 6
ADRENAL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
THYROID FOLLICULAR-CELL ADENOMA FOLLICULAR-CELL CARCINOMA	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	49 2 1
PARATHYROID	-	-	+	+	+	+	-	+	-	-	+	+	-	+	+	+	-	-	-	-	-	+	+	-	+	24
REPRODUCTIVE SYSTEM																										
MAMMARY GLAND ADENOMA, NOS	+	+	+	+	+	+	+	+	+	N	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	50N 1
UTERUS	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
OVARY TERATOMA, NOS	+	+	+	+	+ X	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+	48 1
NERVOUS SYSTEM																										
BRAIN	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
SPECIAL SENSE ORGANS																										
HARDERIAN GLAND ADENOMA, NOS	N	N	N	N	N	N	N	N	N X	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	50N 1
BODY CAVITIES																										
PERITONEUM HEMANGIOSARCOMA	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	N	50N 1
ALL OTHER SYSTEMS																										
MULTIPLE ORGANS NOS SARCOMA, NOS MALIGNANT LYMPHOMA, MIXED TYPE	N	N	N	N	N	N	N	N	N	N	N	N X	N	N	N	N	N	N	N	N	N	N	N X	N	N	50N 1 6

N ANIMALS NECROPSIED

APPENDIX C

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN RATS IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

TABLE C1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
EPIDERMAL INCLUSION CYST	1 (2%)		
STEATITIS	1 (2%)		
INFLAMMATION, CHRONIC	1 (2%)		1 (2%)
FIBROSIS	1 (2%)		
*SUBCUTANEOUS TISSUE	(50)	(50)	(50)
HEMORRHAGE		1 (2%)	
HEMORRHAGIC CYST			1 (2%)
INFLAMMATION, CHRONIC	1 (2%)		
FIBROSIS			2 (4%)
RESPIRATORY SYSTEM			
*NASAL CAVITY	(50)	(50)	(50)
INFLAMMATION, ACUTE SUPPURATIVE			1 (2%)
#TRACHEA	(50)	(50)	(50)
INFLAMMATION, ACUTE SUPPURATIVE			1 (2%)
#LUNG/BRONCHIOLE	(50)	(50)	(50)
HYPERPLASIA, EPITHELIAL			1 (2%)
#LUNG	(50)	(50)	(50)
ASPIRATION, FOREIGN BODY			2 (4%)
FOREIGN BODY, NOS	1 (2%)		
CONGESTION, NOS	4 (8%)	4 (8%)	2 (4%)
EDEMA, NOS		1 (2%)	
PNEUMONIA, ASPIRATION	1 (2%)		1 (2%)
BRONCHOPNEUMONIA, ACUTE		1 (2%)	1 (2%)
INFLAMMATION, FOCAL GRANULOMATOUS	1 (2%)		
PIGMENTATION, NOS	1 (2%)		1 (2%)
HYPERPLASIA, ADENOMATOUS	2 (4%)	1 (2%)	2 (4%)
HYPERPLASIA, ALVEOLAR EPITHELIUM	1 (2%)		1 (2%)
#LUNG/ALVEOLI	(50)	(50)	(50)
HEMORRHAGE		1 (2%)	
HEMATOPOIETIC SYSTEM			
#SPLEEN	(50)	(49)	(50)
ACCESSORY STRUCTURE	1 (2%)		
CONGESTION, NOS	1 (2%)		
FIBROSIS		1 (2%)	
FIBROSIS, FOCAL	2 (4%)		
NECROSIS, ISCHEMIC	1 (2%)	1 (2%)	
PIGMENTATION, NOS			1 (2%)
HEMOSIDEROSIS		1 (2%)	3 (6%)
ATROPHY, FOCAL			1 (2%)
HEMATOPOIESIS	1 (2%)	1 (2%)	1 (2%)
#MANDIBULAR L. NODE	(50)	(50)	(49)
HEMORRHAGE	1 (2%)		
HYPERPLASIA, LYMPHOID			1 (2%)
#CERVICAL LYMPH NODE	(50)	(50)	(49)
HYPERPLASIA, NOS	1 (2%)		
#BRONCHIAL LYMPH NODE	(50)	(50)	(49)
EDEMA, NOS		1 (2%)	
HYPERPLASIA, LYMPHOID		1 (2%)	
#INGUINAL LYMPH NODE	(50)	(50)	(49)
HYPERPLASIA, LYMPHOID	1 (2%)		
#LIVER	(50)	(50)	(50)
HEMATOPOIESIS			1 (2%)

TABLE C1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
HEMATOPOIETIC SYSTEM (Continued)			
#PEYER'S PATCH	(50)	(49)	(49)
HYPERPLASIA, LYMPHOID	2 (4%)		
#COLON	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID	1 (2%)		
#THYMUS	(50)	(50)	(49)
METAPLASIA, OSSEOUS	1 (2%)		
CIRCULATORY SYSTEM			
#MANDIBULAR L. NODE	(50)	(50)	(49)
LYMPHANGIECTASIS			2 (4%)
#MESENTERIC L. NODE	(50)	(50)	(49)
LYMPHANGIECTASIS	1 (2%)		
#HEART	(50)	(50)	(50)
FIBROSIS, FOCAL	1 (2%)	3 (6%)	5 (10%)
FIBROSIS, MULTIFOCAL			1 (2%)
#HEART/ATRIUM	(50)	(50)	(50)
THROMBUS, MURAL	1 (2%)		
#MYOCARDIUM	(50)	(50)	(50)
INFLAMMATION, CHRONIC	39 (78%)	43 (86%)	36 (72%)
FIBROSIS, DIFFUSE	2 (4%)		
*MESENTERIC ARTERY	(50)	(50)	(50)
INFLAMMATION, CHRONIC	1 (2%)		
#PANCREAS	(50)	(49)	(50)
PERIARTERITIS		1 (2%)	
*MESENTERY	(50)	(50)	(50)
PERIARTERITIS	1 (2%)	1 (2%)	
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(49)	(50)	(48)
EDEMA, NOS			1 (2%)
#LIVER	(50)	(50)	(50)
CONGESTION, NOS		1 (2%)	
HEMORRHAGE	1 (2%)		
INFLAMMATION, ACUTE SUPPURATIVE		1 (2%)	
NECROSIS, COAGULATIVE		1 (2%)	
NECROSIS, ZONAL			1 (2%)
CYTOPLASMIC VACUOLIZATION	9 (18%)	7 (14%)	4 (8%)
FOCAL CELLULAR CHANGE	2 (4%)		3 (6%)
HYPERPLASIA, NODULAR	1 (2%)		
ANGIECTASIS		1 (2%)	
#BILE DUCT	(50)	(50)	(50)
INFLAMMATION, CHRONIC			1 (2%)
HYPERPLASIA, NOS	23 (46%)	21 (42%)	15 (30%)
#PANCREAS	(50)	(49)	(50)
ECTOPIA	1 (2%)		
LYMPHOCYTIC INFLAMMATORY INFILTR			1 (2%)
INFLAMMATION, CHRONIC		1 (2%)	
ATROPHY, NOS			1 (2%)
ATROPHY, FOCAL		1 (2%)	
HYPERPLASIA, FOCAL	1 (2%)		
#PANCREATIC ACINUS	(50)	(49)	(50)
ATROPHY, NOS	4 (8%)	5 (10%)	1 (2%)
ATROPHY, FOCAL	3 (6%)		1 (2%)
HYPERPLASIA, NOS	7 (14%)	9 (18%)	8 (16%)
HYPERPLASIA, FOCAL	5 (10%)	5 (10%)	1 (2%)
HYPERPLASIA, DIFFUSE			2 (4%)
#STOMACH	(50)	(50)	(50)
INFLAMMATION, ACUTE SUPPURATIVE		1 (2%)	
HYPERPLASIA, PAPILLARY			1 (2%)

TABLE C1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
DIGESTIVE SYSTEM (Continued)			
#GASTRIC SUBMUCOSA	(50)	(50)	(50)
INFLAMMATION, CHRONIC	1 (2%)		
#FORESTOMACH	(50)	(50)	(50)
EDEMA, NOS		2 (4%)	
ULCER, NOS		2 (4%)	
INFLAMMATION, ACUTE		1 (2%)	
INFLAMMATION, ACUTE/CHRONIC		2 (4%)	
DEGENERATION, NOS		1 (2%)	
HYPERPLASIA, EPITHELIAL		5 (10%)	
HYPERPLASIA, PAPILLARY		1 (2%)	
HYPERKERATOSIS		5 (10%)	
#SMALL INTESTINE	(50)	(49)	(49)
HYPERPLASIA, EPITHELIAL		1 (2%)	
#DUODENUM	(50)	(49)	(49)
INFLAMMATION, ACUTE/CHRONIC			1 (2%)
URINARY SYSTEM			
#KIDNEY	(50)	(50)	(50)
CALCULUS,UNKN GROSS OR MICRO		1 (2%)	
HYDRONEPHROSIS	1 (2%)		1 (2%)
CYST, NOS	1 (2%)		
INFLAMMATION, SUPPURATIVE		1 (2%)	
INFLAMMATION, CHRONIC		1 (2%)	
SCAR	1 (2%)		
NEPHROPATHY	39 (78%)	48 (96%)	39 (78%)
NEPHROSIS, NOS			1 (2%)
NECROSIS, MEDULLARY		1 (2%)	
PIGMENTATION, NOS		1 (2%)	
#KIDNEY/CORTEX	(50)	(50)	(50)
CYST, NOS		1 (2%)	
#KIDNEY/TUBULE	(50)	(50)	(50)
DILATATION, NOS		1 (2%)	
PIGMENTATION, NOS		3 (6%)	9 (18%)
#KIDNEY/PELVIS	(50)	(50)	(50)
HEMORRHAGE	1 (2%)		
#URINARY BLADDER	(50)	(50)	(50)
HEMORRHAGE	1 (2%)		
#U. BLADDER/MUCOSA	(50)	(50)	(50)
HYPERPLASIA, EPITHELIAL		1 (2%)	
#U.BLADDER/SUBMUCOSA	(50)	(50)	(50)
HEMORRHAGE		1 (2%)	
#U.BLADDER/SEROSA	(50)	(50)	(50)
INFLAMMATION, CHRONIC		1 (2%)	
ENDOCRINE SYSTEM			
#PITUITARY	(50)	(48)	(49)
CYST, NOS			2 (4%)
HEMOSIDEROSIS	2 (4%)		
HYPERPLASIA, NOS		1 (2%)	
HYPERPLASIA, FOCAL	5 (10%)	6 (13%)	3 (6%)
ANGIECTASIS	11 (22%)	12 (25%)	6 (12%)
#PITUITARY ACIDOPHIL	(50)	(48)	(49)
HYPERPLASIA, NOS		1 (2%)	
#ADRENAL	(49)	(50)	(50)
CYTOPLASMIC VACUOLIZATION	1 (2%)		
ANGIECTASIS		3 (6%)	1 (2%)

TABLE C1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ENDOCRINE SYSTEM (Continued)			
#ADRENAL CORTEX	(49)	(50)	(50)
ACCESSORY STRUCTURE		1 (2%)	
CYST, NOS			1 (2%)
HEMORRHAGE, CHRONIC		1 (2%)	
DEGENERATION, LIPOID		1 (2%)	
PIGMENTATION, NOS		1 (2%)	4 (8%)
CYTOPLASMIC VACUOLIZATION	2 (4%)	7 (14%)	8 (16%)
FOCAL CELLULAR CHANGE	3 (6%)	1 (2%)	1 (2%)
CYTOLOGIC ALTERATION, NOS		1 (2%)	
HYPERPLASIA, FOCAL		1 (2%)	
ANGIECTASIS		1 (2%)	
#ADRENAL MEDULLA	(49)	(50)	(50)
HYPERPLASIA, FOCAL	1 (2%)	8 (16%)	2 (4%)
HYPERPLASIA, DIFFUSE		1 (2%)	
ANGIECTASIS	1 (2%)		
#THYROID	(49)	(49)	(50)
THYROGLOSSAL DUCT CYST	2 (4%)	1 (2%)	1 (2%)
CYSTIC FOLLICLES	1 (2%)	1 (2%)	
PIGMENTATION, NOS		1 (2%)	5 (10%)
HYPERPLASIA, CYSTIC			1 (2%)
HYPERPLASIA, C-CELL	6 (12%)	7 (14%)	3 (6%)
#THYROID FOLLICLE	(49)	(49)	(50)
PIGMENTATION, NOS		1 (2%)	5 (10%)
HYPERTROPHY, NOS			1 (2%)
HYPERPLASIA, CYSTIC	1 (2%)		
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(50)	(50)	(50)
CYSTIC DUCTS	8 (16%)	6 (12%)	4 (8%)
HYPERPLASIA, CYSTIC			1 (2%)
ADENOSIS	1 (2%)		
*MAMMARY DUCT	(50)	(50)	(50)
HEMORRHAGE		1 (2%)	
*PREPUTIAL GLAND	(50)	(50)	(50)
CYSTIC DUCTS	2 (4%)	2 (4%)	1 (2%)
INFLAMMATION, ACUTE SUPPURATIVE			1 (2%)
INFLAMMATION, CHRONIC		2 (4%)	1 (2%)
INFLAMMATION, CHRONIC SUPPURATIVE	1 (2%)		
ATROPHY, NOS			1 (2%)
HYPERPLASIA, NOS	1 (2%)		
#PROSTATE	(50)	(50)	(50)
DILATATION, NOS		1 (2%)	
CYSTIC DUCTS		1 (2%)	
EDEMA, NOS		1 (2%)	
HEMORRHAGE		1 (2%)	1 (2%)
INFLAMMATION, INTERSTITIAL			1 (2%)
INFLAMMATION, SUPPURATIVE	4 (8%)	6 (12%)	3 (6%)
INFLAMMATION, ACUTE SUPPURATIVE	1 (2%)		
INFLAMMATION, ACUTE/CHRONIC	2 (4%)		1 (2%)
INFLAMMATION, CHRONIC	1 (2%)	4 (8%)	2 (4%)
INFLAMMATION, CHRONIC SUPPURATIVE	1 (2%)	2 (4%)	3 (6%)
NECROSIS, NOS		1 (2%)	
PIGMENTATION, NOS		1 (2%)	
HYPERPLASIA, EPITHELIAL	1 (2%)	2 (4%)	2 (4%)
*SEMINAL VESICLE	(50)	(50)	(50)
HEMORRHAGE			1 (2%)
INFLAMMATION, CHRONIC		2 (4%)	
INFLAMMATION, CHRONIC SUPPURATIVE			1 (2%)
ATROPHY, NOS	1 (2%)	12 (24%)	13 (26%)
HYPERPLASIA, EPITHELIAL		1 (2%)	1 (2%)

TABLE C1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
REPRODUCTIVE SYSTEM (Continued)			
#TESTIS	(50)	(50)	(50)
HEMORRHAGIC CYST			1 (2%)
FIBROSIS			1 (2%)
HEMOSIDEROSIS			1 (2%)
ATROPHY, NOS	5 (10%)	4 (8%)	8 (16%)
ASPERMATOGENESIS	1 (2%)		
HYPERPLASIA, INTERSTITIAL CELL	3 (6%)	1 (2%)	3 (6%)
*SPERMATIC CORD	(50)	(50)	(50)
STEATITIS		1 (2%)	
NERVOUS SYSTEM			
#BRAIN	(50)	(50)	(50)
HEMORRHAGE	3 (6%)		1 (2%)
NECROSIS, NOS	1 (2%)		1 (2%)
#MEDULLA OBLONGATA	(50)	(50)	(50)
GLIOSIS			1 (2%)
*SPINAL NERVE	(50)	(50)	(50)
LYMPHOCYTIC INFLAMMATORY INFILTR		1 (2%)	
SPECIAL SENSE ORGANS			
*EYE	(50)	(50)	(50)
HEMORRHAGE	1 (2%)		1 (2%)
RETINOPATHY	2 (4%)		20 (40%)
CATARACT	2 (4%)		19 (38%)
PHTHISIS BULBI			2 (4%)
*MIDDLE EAR	(50)	(50)	(50)
INFLAMMATION, CHRONIC SUPPURATIVE			1 (2%)
MUSCULOSKELETAL SYSTEM			
*SKULL	(50)	(50)	(50)
HYPEROSTOSIS		1 (2%)	
BODY CAVITIES			
*MEDIASTINUM	(50)	(50)	(50)
HEMORRHAGE	1 (2%)		
*MEDIASTINAL PLEURA	(50)	(50)	(50)
INFLAMMATION, CHRONIC	1 (2%)		
*MESENTERY	(50)	(50)	(50)
STEATITIS	5 (10%)	4 (8%)	5 (10%)
NECROSIS, FAT			1 (2%)
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(50)	(50)	(50)
PIGMENTATION, NOS		44 (88%)	39 (78%)
OMENTUM			
STEATITIS	1		
SPECIAL MORPHOLOGY SUMMARY			
NONE			

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

TABLE C2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
ULCER, NOS		2 (4%)	
INFLAMMATION, CHRONIC		1 (2%)	
HYPERKERATOSIS	1 (2%)	1 (2%)	1 (2%)
*SUBCUT TISSUE	(50)	(50)	(50)
EDEMA, NOS	1 (2%)		
INFLAMMATION, NOS	1 (2%)		
INFLAMMATION, CHRONIC	1 (2%)	1 (2%)	
RESPIRATORY SYSTEM			
*NASAL CAVITY	(50)	(50)	(50)
INFLAMMATION, SUPPURATIVE			1 (2%)
#LUNG	(50)	(50)	(50)
CONGESTION, NOS		1 (2%)	
INFLAMMATION, FOCAL GRANULOMATOUS	1 (2%)		
PIGMENTATION, NOS	4 (8%)		
HYPERPLASIA, ADENOMATOUS	2 (4%)	2 (4%)	1 (2%)
HYPERPLASIA, ALVEOLAR EPITHELIUM		1 (2%)	
METAPLASIA, SQUAMOUS		1 (2%)	
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
LEUKOCYTOSIS, NOS		1 (2%)	
#BONE MARROW	(49)	(50)	(50)
MYELOFIBROSIS	1 (2%)		
HYPERPLASIA, RETICULUM CELL	1 (2%)		
#SPLEEN	(50)	(50)	(50)
CONGESTION, NOS		1 (2%)	
HEMOSIDEROSIS		1 (2%)	4 (8%)
HEMATOPOIESIS	1 (2%)	1 (2%)	2 (4%)
#SPLENIC CAPSULE	(50)	(50)	(50)
FIBROSIS, FOCAL		1 (2%)	
#PANCREATIC L. NODE	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID			1 (2%)
CIRCULATORY SYSTEM			
#MANDIBULAR L. NODE	(50)	(50)	(50)
LYMPHANGIECTASIS	1 (2%)		
#HEART	(50)	(50)	(50)
FIBROSIS, FOCAL	1 (2%)		1 (2%)
#MYOCARDIUM	(50)	(50)	(50)
INFLAMMATION, CHRONIC	28 (56%)	36 (72%)	33 (66%)
INFLAMMATION, CHRONIC FOCAL	1 (2%)	1 (2%)	
#UTERUS	(50)	(50)	(50)
THROMBUS, ORGANIZED		1 (2%)	
#ADRENAL	(50)	(50)	(50)
THROMBOSIS, NOS		1 (2%)	
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(50)	(49)	(50)
INFLAMMATION, ACUTE SUPPURATIVE			1 (2%)

TABLE C2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
DIGESTIVE SYSTEM (Continued)			
#LIVER	(50)	(50)	(50)
HEMORRHAGE	1 (2%)		
LYMPHOCYTIC INFLAMMATORY INFILTR	2 (4%)		2 (4%)
INFLAMMATION, ACUTE/CHRONIC			1 (2%)
INFLAMMATION, FOCAL GRANULOMATOUS	1 (2%)	3 (6%)	1 (2%)
FIBROSIS, FOCAL			1 (2%)
NECROSIS, NOS	1 (2%)		
NECROSIS, ZONAL			2 (4%)
PIGMENTATION, NOS	1 (2%)		
HEMOSIDEROSIS			1 (2%)
CYTOPLASMIC VACUOLIZATION	4 (8%)	2 (4%)	
BASOPHILIC CYTO CHANGE	1 (2%)		
FOCAL CELLULAR CHANGE	1 (2%)	4 (8%)	
#LIVER/CENTRILOBULAR	(50)	(50)	(50)
CYTOPLASMIC VACUOLIZATION		1 (2%)	
#BILE DUCT	(50)	(50)	(50)
INFLAMMATION, CHRONIC			1 (2%)
HYPERPLASIA, NOS	4 (8%)		
HYPERPLASIA, FOCAL			1 (2%)
#PANCREAS	(50)	(50)	(50)
ATROPHY, FOCAL		1 (2%)	
#PANCREATIC ACINUS	(50)	(50)	(50)
ATROPHY, NOS	1 (2%)	4 (8%)	1 (2%)
ATROPHY, FOCAL	2 (4%)	1 (2%)	
HYPERPLASIA, NOS		2 (4%)	1 (2%)
HYPERPLASIA, FOCAL	1 (2%)	2 (4%)	
HYPERPLASIA, DIFFUSE		1 (2%)	
#ESOPHAGUS	(50)	(50)	(50)
HYPERKERATOSIS		1 (2%)	
#GASTRIC SUBMUCOSA	(50)	(50)	(50)
EDEMA, NOS	1 (2%)		
INFLAMMATION, CHRONIC	1 (2%)		
#FORESTOMACH	(50)	(50)	(50)
ULCER, NOS		1 (2%)	
INFLAMMATION, CHRONIC	1 (2%)		
HYPERPLASIA, EPITHELIAL	1 (2%)	1 (2%)	
HYPERPLASIA, PAPILLARY			1 (2%)
HYPERKERATOSIS	1 (2%)	1 (2%)	
#JEJUNUM	(50)	(49)	(49)
INFLAMMATION, CHRONIC FOCAL		1 (2%)	
*RECTUM	(50)	(50)	(50)
PARASITISM		1 (2%)	
URINARY SYSTEM			
#KIDNEY	(50)	(50)	(50)
CYST, NOS		1 (2%)	1 (2%)
LYMPHOCYTIC INFLAMMATORY INFILTR	1 (2%)		
SCAR	1 (2%)		
NEPHROPATHY	7 (14%)	12 (24%)	20 (40%)
PIGMENTATION, NOS	1 (2%)		
ATROPHY, NOS	1 (2%)		
#KIDNEY/TUBULE	(50)	(50)	(50)
CALCIFICATION, FOCAL	1 (2%)		
PIGMENTATION, NOS	2 (4%)	2 (4%)	4 (8%)
#KIDNEY/PELVIS	(50)	(50)	(50)
MINERALIZATION		1 (2%)	
HYPERPLASIA, EPITHELIAL		1 (2%)	
#URINARY BLADDER	(50)	(50)	(50)
HYPERPLASIA, EPITHELIAL			1 (2%)

TABLE C2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
URINARY SYSTEM (Continued)			
#U. BLADDER/SUBMUCOSA	(50)	(50)	(50)
INFLAMMATION, CHRONIC			1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(50)	(50)	(50)
CYST, NOS	3 (6%)	5 (10%)	4 (8%)
MULTIPLE CYSTS	1 (2%)		2 (4%)
HEMORRHAGE			1 (2%)
HEMORRHAGIC CYST			1 (2%)
NECROSIS, NOS			1 (2%)
HEMOSIDEROSIS		2 (4%)	1 (2%)
HYPERPLASIA, NOS	1 (2%)	2 (4%)	
HYPERPLASIA, FOCAL	3 (6%)	3 (6%)	1 (2%)
ANGIECTASIS	15 (30%)	16 (32%)	15 (30%)
#ADRENAL	(50)	(50)	(50)
ACCESSORY STRUCTURE	1 (2%)		
PIGMENTATION, NOS	1 (2%)		
CYTOPLASMIC VACUOLIZATION	2 (4%)	1 (2%)	
ANGIECTASIS		1 (2%)	
#ADRENAL CORTEX	(50)	(50)	(50)
ACCESSORY STRUCTURE		1 (2%)	
CYST, NOS	1 (2%)	1 (2%)	
DEGENERATION, NOS			1 (2%)
CYTOPLASMIC VACUOLIZATION	8 (16%)	11 (22%)	10 (20%)
FOCAL CELLULAR CHANGE	2 (4%)	2 (4%)	1 (2%)
HYPERPLASIA, NODULAR	1 (2%)		
HYPERPLASIA, FOCAL		1 (2%)	
ANGIECTASIS		1 (2%)	1 (2%)
#ADRENAL MEDULLA	(50)	(50)	(50)
HYPERPLASIA, FOCAL			1 (2%)
ANGIECTASIS			1 (2%)
#THYROID	(50)	(50)	(50)
THYROGLOSSAL DUCT CYST	1 (2%)	5 (10%)	2 (4%)
CYSTIC FOLLICLES	2 (4%)		1 (2%)
PIGMENTATION, NOS			3 (6%)
HYPERPLASIA, CYSTIC			1 (2%)
HYPERPLASIA, C-CELL	8 (16%)	9 (18%)	3 (6%)
HYPERPLASIA, FOLLICULAR-CELL		1 (2%)	1 (2%)
#THYROID FOLLICLE	(50)	(50)	(50)
PIGMENTATION, NOS		2 (4%)	1 (2%)
HYPERPLASIA, CYSTIC		1 (2%)	2 (4%)
#PARATHYROID	(50)	(48)	(48)
ANGIECTASIS		1 (2%)	
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(50)	(50)	(50)
CYSTIC DUCTS	23 (46%)	25 (50%)	13 (26%)
HYPERPLASIA, CYSTIC	1 (2%)	1 (2%)	1 (2%)
ADENOSIS		1 (2%)	
*MAMMARY LOBULE	(50)	(50)	(50)
HYPERPLASIA, NOS	1 (2%)		1 (2%)
*CLITORAL GLAND	(50)	(50)	(50)
CYSTIC DUCTS		2 (4%)	1 (2%)
INFLAMMATION, ACUTE SUPPURATIVE		1 (2%)	
INFLAMMATION, CHRONIC			1 (2%)
INFLAMMATION, CHRONIC SUPPURATIVE	1 (2%)	2 (4%)	
#UTERUS	(50)	(50)	(50)
HYDROMETRA		1 (2%)	1 (2%)
HEMORRHAGE	2 (4%)	1 (2%)	
HEMORRHAGE, CHRONIC			1 (2%)

TABLE C2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
REPRODUCTIVE SYSTEM (Continued)			
#UTERUS/ENDOMETRIUM	(50)	(50)	(50)
CYST, NOS		1 (2%)	2 (4%)
INFLAMMATION, SUPPURATIVE			1 (2%)
HYPERPLASIA, NOS	2 (4%)	2 (4%)	2 (4%)
HYPERPLASIA, CYSTIC	10 (20%)	17 (34%)	10 (20%)
HYPERPLASIA, ADENOMATOUS		3 (6%)	1 (2%)
#OVARY	(50)	(50)	(50)
CYST, NOS		1 (2%)	1 (2%)
CYSTIC FOLLICLES	3 (6%)	2 (4%)	2 (4%)
NERVOUS SYSTEM			
#BRAIN	(50)	(50)	(50)
HEMORRHAGE	2 (4%)		
NECROSIS, NOS	1 (2%)		
#CEREBELLUM	(50)	(50)	(50)
STATUS SPONGIOSUS		1 (2%)	
SPECIAL SENSE ORGANS			
*EYE	(50)	(50)	(50)
RETINOPATHY	1 (2%)	19 (38%)	
CATARACT	1 (2%)	19 (38%)	
*EYE/CORNEA	(50)	(50)	(50)
INFLAMMATION, CHRONIC			1 (2%)
MUSCULOSKELETAL SYSTEM			
*SKULL	(50)	(50)	(50)
HYPEROSTOSIS			1 (2%)
*TARSAL JOINT	(50)	(50)	(50)
OSTEOARTHRITIS		1 (2%)	
BODY CAVITIES			
*MESENTERY	(50)	(50)	(50)
STEATITIS	3 (6%)	2 (4%)	
NECROSIS, FAT			1 (2%)
PIGMENTATION, NOS		1 (2%)	
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(50)	(50)	(50)
PIGMENTATION, NOS		46 (92%)	44 (88%)
OMENTUM			
STEATITIS	1		1
BROAD LIGAMENT			
STEATITIS	4	3	3
INFLAMMATION, ACUTE/CHRONIC			1
SPECIAL MORPHOLOGY SUMMARY			
NONE			

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

APPENDIX D

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

TABLE D1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
ULCER, NOS		1 (2%)	
INFLAMMATION, CHRONIC		1 (2%)	1 (2%)
ULCER, CHRONIC		2 (4%)	1 (2%)
FIBROSIS		1 (2%)	
*SUBCUT TISSUE	(50)	(50)	(50)
INFLAMMATION, FOCAL	1 (2%)		
INFLAMMATION, GRANULOMATOUS	1 (2%)		
INFECTION, FUNGAL	1 (2%)		
RESPIRATORY SYSTEM			
*NASAL MUCOSA	(50)	(50)	(50)
INFLAMMATION, NOS	1 (2%)		
#LUNG	(50)	(49)	(50)
CONGESTION, NOS	4 (8%)	10 (20%)	1 (2%)
INFLAMMATION, FOCAL		1 (2%)	5 (10%)
PNEUMONIA, ASPIRATION			1 (2%)
INFLAMMATION, SUPPURATIVE			1 (2%)
INFLAMMATION, CHRONIC FOCAL	1 (2%)		
HYPERPLASIA, ALVEOLAR EPITHELIUM		1 (2%)	1 (2%)
HISTIOCYTOSIS		2 (4%)	
#LUNG/ALVEOLI	(50)	(49)	(50)
HISTIOCYTOSIS	4 (8%)	1 (2%)	1 (2%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID	2 (4%)	3 (6%)	1 (2%)
HEMATOPOIESIS	1 (2%)		1 (2%)
#BONE MARROW	(50)	(49)	(50)
HYPERPLASIA, GRANULOCYTIC		2 (4%)	2 (4%)
#SPLEEN	(50)	(50)	(50)
ATROPHY, NOS			1 (2%)
ANGIECTASIS		1 (2%)	1 (2%)
HYPERPLASIA, LYMPHOID	1 (2%)	2 (4%)	3 (6%)
HEMATOPOIESIS	9 (18%)	5 (10%)	6 (12%)
#MANDIBULAR L. NODE	(50)	(49)	(50)
CONGESTION, NOS	1 (2%)		
HYPERPLASIA, NOS	1 (2%)	2 (4%)	2 (4%)
#MESENTERIC L. NODE	(50)	(49)	(50)
CONGESTION, NOS	16 (32%)	7 (14%)	4 (8%)
ANGIECTASIS	10 (20%)	13 (27%)	16 (32%)
HYPERPLASIA, LYMPHOID	2 (4%)	1 (2%)	
HEMATOPOIESIS	1 (2%)		
#INGUINAL LYMPH NODE	(50)	(49)	(50)
HYPERPLASIA, NOS	1 (2%)	1 (2%)	
HYPERPLASIA, LYMPHOID		2 (4%)	
#LUNG	(50)	(49)	(50)
LEUKOCYTOSIS, NOS		2 (4%)	1 (2%)
#LIVER	(50)	(50)	(50)
LEUKOCYTOSIS, NOS	1 (2%)	1 (2%)	1 (2%)
HEMATOPOIESIS			1 (2%)
*GALLBLADDER	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID		1 (2%)	
#ILEUM	(50)	(48)	(50)
HYPERPLASIA, LYMPHOID		1 (2%)	

TABLE D1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
HEMATOPOIETIC SYSTEM (Continued)			
*RECTUM	(50)	(50)	(50)
HYPERPLASIA, RETICULUM CELL		1 (2%)	
#KIDNEY	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID	3 (6%)	6 (12%)	1 (2%)
MASTOCYTOSIS		1 (2%)	
CIRCULATORY SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
PERIARTERITIS	1 (2%)	1 (2%)	
#INGUINAL LYMPH NODE	(50)	(49)	(50)
LYMPHANGIECTASIS		1 (2%)	
#MYOCARDIUM	(50)	(49)	(50)
ANGIECTASIS		1 (2%)	
#LIVER	(50)	(50)	(50)
THROMBOSIS, NOS		1 (2%)	1 (2%)
DIGESTIVE SYSTEM			
#LIVER	(50)	(50)	(50)
DEFORMITY, NOS	1 (2%)		
CYST, NOS		1 (2%)	
HEMORRHAGE	2 (4%)		
INFLAMMATION, FOCAL		1 (2%)	1 (2%)
INFLAMMATION, CHRONIC FOCAL	1 (2%)		
FIBROSIS, FOCAL		1 (2%)	
NECROSIS, FOCAL	1 (2%)	2 (4%)	
INFARCT, NOS		1 (2%)	1 (2%)
METAMORPHOSIS FATTY	2 (4%)		2 (4%)
PIGMENTATION, NOS	3 (6%)		1 (2%)
FOCAL CELLULAR CHANGE	2 (4%)	1 (2%)	2 (4%)
ANGIECTASIS	2 (4%)	1 (2%)	
#LIVER/HEPATOCYTES	(50)	(50)	(50)
NUCLEAR ALTERATION		1 (2%)	
*GALLBLADDER	(50)	(50)	(50)
HYPERPLASIA, ADENOMATOUS		2 (4%)	
*MUCOSA OF GALLBLADDER	(50)	(50)	(50)
CYST, NOS	1 (2%)		
#BILE DUCT	(50)	(50)	(50)
DILATATION, NOS	1 (2%)		
CYST, NOS	1 (2%)		
HYPERPLASIA, NOS	1 (2%)	1 (2%)	1 (2%)
#PANCREAS	(50)	(50)	(50)
CYSTIC DUCTS			2 (4%)
CONGESTION, NOS	1 (2%)		
ATROPHY, NOS			1 (2%)
ATROPHY, FOCAL		1 (2%)	3 (6%)
#GASTRIC FUNDAL GLAND	(50)	(50)	(50)
CYST, NOS		1 (2%)	
#STOMACH WALL	(50)	(50)	(50)
INFLAMMATION, NOS		1 (2%)	
#FORESTOMACH	(50)	(50)	(50)
INFLAMMATION, FOCAL		1 (2%)	
HYPERPLASIA, EPITHELIAL	1 (2%)	3 (6%)	
#COLONIC SUBMUCOSA	(50)	(50)	(50)
EDEMA, NOS	1 (2%)		
#CECUM	(50)	(50)	(50)
ANGIECTASIS	1 (2%)		

TABLE D1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
URINARY SYSTEM			
#KIDNEY	(50)	(50)	(50)
INFLAMMATION, FOCAL			1 (2%)
INFLAMMATION, CHRONIC	1 (2%)		
INFLAMMATION, CHRONIC FOCAL			1 (2%)
NEPHROSIS, NOS	25 (50%)	32 (64%)	27 (54%)
GLOMERULOSCLEROSIS, NOS			1 (2%)
ATROPHY, FOCAL			2 (4%)
#KIDNEY/MEDULLA	(50)	(50)	(50)
CALCIFICATION, NOS			1 (2%)
#BOWMAN'S CAPSULE	(50)	(50)	(50)
BASEMENT MEMBRANE, ALTERATION	2 (4%)		
#KIDNEY/TUBULE	(50)	(50)	(50)
CYST, NOS		2 (4%)	
DEGENERATION, HYALINE		1 (2%)	
CYTOPLASMIC VACUOLIZATION	1 (2%)		
#URINARY BLADDER	(49)	(49)	(50)
HEMORRHAGE			1 (2%)
HYPERPLASIA, EPITHELIAL	1 (2%)		1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(46)	(47)	(44)
EMBRYONAL DUCT CYST	1 (2%)	1 (2%)	
FOCAL CELLULAR CHANGE		1 (2%)	3 (7%)
#ADRENAL	(50)	(50)	(50)
ACCESSORY STRUCTURE	1 (2%)		
#ADRENAL CORTEX	(50)	(50)	(50)
CYST, NOS		1 (2%)	
FIBROSIS, FOCAL		1 (2%)	
HYPERPLASIA, NODULAR	1 (2%)	3 (6%)	
HYPERPLASIA, FOCAL			1 (2%)
#ADRENAL MEDULLA	(50)	(50)	(50)
FIBROSIS, FOCAL			1 (2%)
HYPERPLASIA, FOCAL			1 (2%)
#THYROID	(48)	(50)	(50)
CYSTIC FOLLICLES		1 (2%)	
DEGENERATION, CYSTIC	14 (29%)	15 (30%)	13 (26%)
PIGMENTATION, NOS	1 (2%)	38 (76%)	47 (94%)
HYPERPLASIA, CYSTIC	1 (2%)		1 (2%)
HYPERPLASIA, FOLLICULAR-CELL		1 (2%)	
#THYROID FOLLICLE	(48)	(50)	(50)
CRYSTALS, NOS	3 (6%)	1 (2%)	2 (4%)
HYPERPLASIA, CYSTIC	3 (6%)	9 (18%)	12 (24%)
REPRODUCTIVE SYSTEM			
*PREPUTIAL GLAND	(50)	(50)	(50)
MULTIPLE CYSTS		3 (6%)	
CYSTIC DUCTS	3 (6%)	3 (6%)	5 (10%)
INFLAMMATION, NOS	1 (2%)	2 (4%)	5 (10%)
INFLAMMATION, SUPPURATIVE	1 (2%)	3 (6%)	1 (2%)
DEGENERATION, CYSTIC			1 (2%)
*SEMINAL VESICLE	(50)	(50)	(50)
DILATATION, NOS	2 (4%)	1 (2%)	5 (10%)
*COAGULATING GLAND	(50)	(50)	(50)
DILATATION, NOS	2 (4%)	1 (2%)	4 (8%)
COLLOID CYST			1 (2%)
#TESTIS	(50)	(50)	(50)
ATROPHY, NOS		1 (2%)	1 (2%)
ATROPHY, FOCAL			1 (2%)

TABLE D1. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
REPRODUCTIVE SYSTEM (Continued)			
*EPIDIDYMIS	(50)	(50)	(50)
GRANULOMA, SPERMATIC	1 (2%)	2 (4%)	1 (2%)
*SPERMATIC CORD	(50)	(50)	(50)
NECROSIS, FAT	1 (2%)	1 (2%)	2 (4%)
NERVOUS SYSTEM			
#BRAIN/THALAMUS	(50)	(50)	(50)
PSAMMOMA BODIES	17 (34%)	28 (56%)	20 (40%)
SPECIAL SENSE ORGANS			
*EYE	(50)	(50)	(50)
CATARACT	1 (2%)		1 (2%)
PHTHISIS BULBI			2 (4%)
*EYE/CORNEA	(50)	(50)	(50)
INFLAMMATION, NOS	1 (2%)		
ULCER, NOS			1 (2%)
MUSCULOSKELETAL SYSTEM			
*SKULL	(50)	(50)	(50)
HYPEROSTOSIS	1 (2%)		
BODY CAVITIES			
*THORACIC CAVITY	(50)	(50)	(50)
REACTION, FOREIGN BODY			1 (2%)
*ABDOMINAL WALL	(50)	(50)	(50)
ANGIECTASIS		1 (2%)	
*PERITONEUM	(50)	(50)	(50)
INFLAMMATION, NOS			1 (2%)
*MESENTERY	(50)	(50)	(50)
INFLAMMATION, FOCAL	1 (2%)		
NECROSIS, FAT	2 (4%)	3 (6%)	5 (10%)
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(50)	(50)	(50)
DILATATION, NOS			1 (2%)
INFLAMMATION, SUPPURATIVE			2 (4%)
REACTION, FOREIGN BODY			1 (2%)
OMENTUM			
NECROSIS, FAT			1
CALCIFICATION, NOS			1
SPECIAL MORPHOLOGY SUMMARY			
NONE			

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

TABLE D2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS NECROPSIED	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY	50	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(50)	(50)	(50)
INFLAMMATION, NOS	1 (2%)		
INFLAMMATION, CHRONIC			2 (4%)
RESPIRATORY SYSTEM			
#LUNG	(50)	(49)	(50)
CONGESTION, NOS		2 (4%)	3 (6%)
INFLAMMATION, FOCAL			1 (2%)
INFLAMMATION, INTERSTITIAL	1 (2%)	1 (2%)	
HISTIOCYTOSIS		1 (2%)	1 (2%)
#LUNG/ALVEOLI	(50)	(49)	(50)
HISTIOCYTOSIS		1 (2%)	2 (4%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(50)	(50)	(50)
LEUKOCYTOSIS, NOS	9 (18%)	15 (30%)	9 (18%)
HYPERPLASIA, LYMPHOID	4 (8%)	3 (6%)	5 (10%)
HEMATOPOIESIS	11 (22%)	11 (22%)	9 (18%)
*MEDIASTINUM	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID		1 (2%)	
#BONE MARROW	(50)	(49)	(49)
MYELOFIBROSIS		1 (2%)	
HYPERPLASIA, GRANULOCYTIC	7 (14%)	7 (14%)	5 (10%)
#SPLEEN	(50)	(50)	(50)
PIGMENTATION, NOS			1 (2%)
HYPERPLASIA, LYMPHOID		1 (2%)	
HEMATOPOIESIS	19 (38%)	19 (38%)	19 (38%)
#LYMPH NODE	(50)	(49)	(50)
HYPERPLASIA, NOS	3 (6%)	1 (2%)	
HYPERPLASIA, LYMPHOID			1 (2%)
#MANDIBULAR L. NODE	(50)	(49)	(50)
HYPERPLASIA, NOS			1 (2%)
#MEDIASTINAL L.NODE	(50)	(49)	(50)
HYPERPLASIA, NOS	2 (4%)		1 (2%)
HYPERPLASIA, PLASMA CELL	1 (2%)		
#ABDOMINAL LYMPH NODE	(50)	(49)	(50)
HYPERPLASIA, NOS	1 (2%)	1 (2%)	
#MESENTERIC L. NODE	(50)	(49)	(50)
CONGESTION, NOS			2 (4%)
INFLAMMATION, NOS			1 (2%)
LYMPHOID DEPLETION		1 (2%)	
HYPERPLASIA, NOS		1 (2%)	
ANGIECTASIS	1 (2%)	2 (4%)	
HYPERPLASIA, LYMPHOID		1 (2%)	1 (2%)
#RENAL LYMPH NODE	(50)	(49)	(50)
CONGESTION, NOS			1 (2%)
HYPERPLASIA, NOS	5 (10%)	1 (2%)	5 (10%)
#ILIAC LYMPH NODE	(50)	(49)	(50)
HYPERPLASIA, NOS	2 (4%)	1 (2%)	1 (2%)
#INGUINAL LYMPH NODE	(50)	(49)	(50)
HYPERPLASIA, LYMPHOID	1 (2%)		
#LUNG	(50)	(49)	(50)
LEUKOCYTOSIS, NOS	1 (2%)		3 (6%)
HYPERPLASIA, LYMPHOID		1 (2%)	1 (2%)

TABLE D2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
HEMATOPOIETIC SYSTEM (Continued)			
#LIVER	(50)	(50)	(50)
LEUKOCYTOSIS, NOS	17 (34%)	13 (26%)	11 (22%)
HEMATOPOIESIS		1 (2%)	
#KIDNEY	(50)	(50)	(50)
HYPERPLASIA, LYMPHOID	1 (2%)		
CIRCULATORY SYSTEM			
#LUNG	(50)	(49)	(50)
EMBOLUS, SEPTIC	1 (2%)		
#HEART	(50)	(50)	(50)
INFLAMMATION, FOCAL		1 (2%)	1 (2%)
#MYOCARDIUM	(50)	(50)	(50)
PERIARTERITIS			1 (2%)
#LIVER	(50)	(50)	(50)
THROMBOSIS, NOS	1 (2%)		
*MESENTERY	(50)	(50)	(50)
PERIARTERITIS		1 (2%)	
DIGESTIVE SYSTEM			
#LIVER	(50)	(50)	(50)
NECROSIS, FOCAL	1 (2%)	1 (2%)	1 (2%)
METAMORPHOSIS FATTY		2 (4%)	1 (2%)
PIGMENTATION, NOS	1 (2%)		
CYTOPLASMIC VACUOLIZATION			2 (4%)
ANGIECTASIS			1 (2%)
#LIVER/KUPFFER CELL	(50)	(50)	(50)
PIGMENTATION, NOS			1 (2%)
#LIVER/HEPATOCYTES	(50)	(50)	(50)
NUCLEAR ALTERATION	1 (2%)	1 (2%)	
*GALLBLADDER	(50)	(50)	(50)
EDEMA, NOS	1 (2%)		
#PANCREAS	(50)	(50)	(50)
INFLAMMATION, NOS			1 (2%)
ATROPHY, NOS	1 (2%)		1 (2%)
ATROPHY, FOCAL		1 (2%)	1 (2%)
#STOMACH	(50)	(50)	(48)
INFLAMMATION, NOS		1 (2%)	
#GASTRIC MUCOSA	(50)	(50)	(48)
INFLAMMATION, FOCAL		1 (2%)	
#FORESTOMACH	(50)	(50)	(48)
ULCER, NOS	2 (4%)		
INFLAMMATION, FOCAL	1 (2%)		
HYPERPLASIA, EPITHELIAL	3 (6%)	2 (4%)	1 (2%)
#COLON	(49)	(49)	(49)
INFLAMMATION, NOS			1 (2%)
URINARY SYSTEM			
#KIDNEY	(50)	(50)	(50)
HYDRONEPHROSIS			2 (4%)
INFLAMMATION, FOCAL		1 (2%)	
LYMPHOCYTIC INFLAMMATORY INFILTR			1 (2%)
INFLAMMATION, CHRONIC			1 (2%)
PYELONEPHRITIS, CHRONIC		1 (2%)	
INFLAMMATION, CHRONIC FOCAL			2 (4%)
FIBROSIS, FOCAL			1 (2%)
NEPHROSIS, NOS	1 (2%)	5 (10%)	10 (20%)
GLOMERULOSCLEROSIS, NOS	1 (2%)	1 (2%)	1 (2%)
ATROPHY, NOS		1 (2%)	
ATROPHY, FOCAL			1 (2%)

TABLE D2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
URINARY SYSTEM (Continued)			
#KIDNEY/TUBULE	(50)	(50)	(50)
PIGMENTATION, NOS			1 (2%)
#URINARY BLADDER	(50)	(50)	(49)
INFLAMMATION, NOS		1 (2%)	
HYPERPLASIA, EPITHELIAL		1 (2%)	
#U. BLADDER/MUCOSA	(50)	(50)	(49)
CYTOPLASMIC VACUOLIZATION		1 (2%)	
ENDOCRINE SYSTEM			
#PITUITARY	(47)	(45)	(43)
FOCAL CELLULAR CHANGE	1 (2%)		1 (2%)
HYPERPLASIA, FOCAL	5 (11%)	3 (7%)	1 (2%)
ANGIECTASIS	1 (2%)	1 (2%)	1 (2%)
#ADRENAL	(50)	(49)	(49)
INFLAMMATION, FOCAL			1 (2%)
NECROSIS, FOCAL	1 (2%)		
#ADRENAL/CAPSULE	(50)	(49)	(49)
HYPERPLASIA, FOCAL		1 (2%)	
#ADRENAL CORTEX	(50)	(49)	(49)
INFLAMMATION, FOCAL	1 (2%)		
#THYROID	(49)	(48)	(49)
CYSTIC FOLLICLES		1 (2%)	3 (6%)
INFLAMMATION, FOCAL			4 (8%)
DEGENERATION, CYSTIC	11 (22%)	12 (25%)	11 (22%)
PIGMENTATION, NOS	2 (4%)	13 (27%)	24 (49%)
HYPERPLASIA, CYSTIC			1 (2%)
HYPERPLASIA, FOLLICULAR-CELL		1 (2%)	
#THYROID FOLLICLE	(49)	(48)	(49)
INFLAMMATION, FOCAL	1 (2%)		
HYPERPLASIA, CYSTIC	5 (10%)	5 (10%)	3 (6%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(50)	(50)	(50)
CYSTIC DISEASE	6 (12%)	3 (6%)	8 (16%)
*CLITORAL GLAND	(50)	(50)	(50)
CYSTIC DUCTS		1 (2%)	
*VAGINAL MUCOSA	(50)	(50)	(50)
HYPERPLASIA, EPITHELIAL	1 (2%)		
#UTERUS	(50)	(50)	(50)
HEMATOMA, NOS			1 (2%)
INFLAMMATION, SUPPURATIVE	3 (6%)	2 (4%)	1 (2%)
#UTERUS/ENDOMETRIUM	(50)	(50)	(50)
HYPERPLASIA, CYSTIC	15 (30%)	17 (34%)	18 (36%)
#OVARY	(45)	(48)	(48)
CYST, NOS	1 (2%)	1 (2%)	
FOLLICULAR CYST, NOS	8 (18%)	5 (10%)	6 (13%)
PAROVARIAN CYST			1 (2%)
INFLAMMATION, SUPPURATIVE		1 (2%)	3 (6%)
ABSCESS, CHRONIC		1 (2%)	1 (2%)
HYPERPLASIA, NOS		1 (2%)	
ANGIECTASIS			1 (2%)
NERVOUS SYSTEM			
#BRAIN/THALAMUS	(50)	(50)	(50)
PSAMMOMA BODIES	10 (20%)	17 (34%)	7 (14%)

TABLE D2. SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	CONTROL (VEH)	LOW DOSE	HIGH DOSE
SPECIAL SENSE ORGANS			
NONE			
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*BODY CAVITIES	(50)	(50)	(50)
INFECTION, BACTERIAL			1 (2%)
*ABDOMINAL CAVITY	(50)	(50)	(50)
HEMORRHAGIC CYST			1 (2%)
HEMORRHAGE, CHRONIC		1 (2%)	
*PERITONEUM	(50)	(50)	(50)
INFLAMMATION, NOS			1 (2%)
NECROSIS, FAT			1 (2%)
CALCIFICATION, FOCAL			1 (2%)
*MESENTERY	(50)	(50)	(50)
INFLAMMATION, NOS		1 (2%)	
NECROSIS, FAT	1 (2%)		1 (2%)
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(50)	(50)	(50)
INFLAMMATION, NOS			1 (2%)
INFLAMMATION, SUPPURATIVE	33 (66%)	32 (64%)	29 (58%)
INFLAMMATION, CHRONIC SUPPURATIVE			1 (2%)
AMYLOIDOSIS		1 (2%)	
BROAD LIGAMENT			
NECROSIS, FAT	2	1	1
SPECIAL MORPHOLOGY SUMMARY			
NONE			

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

APPENDIX E

ANALYSES OF PRIMARY TUMORS IN RATS AND MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3

TABLE E1. ANALYSIS OF PRIMARY TUMORS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	Vehicle Control	250 mg/kg	500 mg/kg
Skin: Basal Cell Carcinoma			
Overall Rates (a)	0/50 (0%)	3/50 (6%)	1/50 (2%)
Adjusted Rates (b)	0.0%	8.5%	3.1%
Terminal Rates (c)	0/34 (0%)	2/34 (6%)	1/32 (3%)
Life Table Tests (d)	P=0.355	P=0.121	P=0.488
Incidental Tumor Tests (d)	P=0.293	P=0.138	P=0.488
Cochran-Armitage Trend Test (d)	P=0.378		
Fisher Exact Tests		P=0.121	P=0.500
Skin: Keratoacanthoma			
Overall Rates (a)	2/50 (4%)	3/50 (6%)	2/50 (4%)
Adjusted Rates (b)	5.9%	7.8%	5.3%
Terminal Rates (c)	2/34 (6%)	2/34 (6%)	1/32 (3%)
Life Table Tests (d)	P=0.564	P=0.500	P=0.669
Incidental Tumor Tests (d)	P=0.450N	P=0.561	P=0.603N
Cochran-Armitage Trend Test (d)	P=0.594		
Fisher Exact Tests		P=0.500	P=0.691
Subcutaneous Tissue: Fibroma			
Overall Rates (a)	3/50 (6%)	7/50 (14%)	2/50 (4%)
Adjusted Rates (b)	8.8%	18.4%	6.3%
Terminal Rates (c)	3/34 (9%)	5/34 (15%)	2/32 (6%)
Life Table Tests (d)	P=0.467N	P=0.164	P=0.528N
Incidental Tumor Tests (d)	P=0.394N	P=0.138	P=0.528N
Cochran-Armitage Trend Test (d)	P=0.427N		
Fisher Exact Tests		P=0.159	P=0.500N
Integumentary System: Fibroma			
Overall Rates (a)	4/50 (8%)	7/50 (14%)	2/50 (4%)
Adjusted Rates (b)	11.8%	18.4%	6.3%
Terminal Rates (c)	4/34 (12%)	5/34 (15%)	2/32 (6%)
Life Table Tests (d)	P=0.334N	P=0.266	P=0.364N
Incidental Tumor Tests (d)	P=0.272N	P=0.236	P=0.364N
Cochran-Armitage Trend Test (d)	P=0.297N		
Fisher Exact Tests		P=0.262	P=0.339N
Integumentary System: Fibroma or Neurofibrosarcoma			
Overall Rates (a)	5/50 (10%)	7/50 (14%)	2/50 (4%)
Adjusted Rates (b)	14.7%	18.4%	6.3%
Terminal Rates (c)	5/34 (15%)	5/34 (15%)	2/32 (6%)
Life Table Tests (d)	P=0.225N	P=0.384	P=0.239N
Incidental Tumor Tests (d)	P=0.177N	P=0.351	P=0.239N
Cochran-Armitage Trend Test (d)	P=0.195N		
Fisher Exact Tests		P=0.380	P=0.218N
Hematopoietic System: Mononuclear Cell Leukemia			
Overall Rates (a)	9/50 (18%)	3/50 (6%)	3/50 (6%)
Adjusted Rates (b)	22.2%	7.9%	8.2%
Terminal Rates (c)	4/34 (12%)	1/34 (3%)	1/32 (3%)
Life Table Tests (d)	P=0.057N	P=0.075N	P=0.105N
Incidental Tumor Tests (d)	P=0.081N	P=0.072N	P=0.149N
Cochran-Armitage Trend Test (d)	P=0.033N		
Fisher Exact Tests		P=0.061N	P=0.061N
Liver: Neoplastic Nodule			
Overall Rates (a)	3/50 (6%)	0/50 (0%)	1/50 (2%)
Adjusted Rates (b)	8.8%	0.0%	3.1%
Terminal Rates (c)	3/34 (9%)	0/34 (0%)	1/32 (3%)
Life Table Tests (d)	P=0.187N	P=0.121N	P=0.326N
Incidental Tumor Tests (d)	P=0.187N	P=0.121N	P=0.326N
Cochran-Armitage Trend Test (d)	P=0.176N		
Fisher Exact Tests		P=0.121N	P=0.309N

TABLE E1. ANALYSIS OF PRIMARY TUMORS IN MALE RATS IN THE TWO- YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	250 mg/kg	500 mg/kg
Liver: Neoplastic Nodule or Hepatocellular Carcinoma			
Overall Rates (a)	4/50 (8%)	1/50 (2%)	3/50 (6%)
Adjusted Rates (b)	11.0%	2.9%	9.4%
Terminal Rates (c)	3/34 (9%)	1/34 (3%)	3/32 (9%)
Life Table Tests (d)	P=0.448N	P=0.181N	P=0.545N
Incidental Tumor Tests (d)	P=0.485N	P=0.166N	P=0.616N
Cochran-Armitage Trend Test (d)	P=0.412N		
Fisher Exact Tests		P=0.181N	P=0.500N
Pancreas: Acinar Cell Adenoma			
Overall Rates (a)	11/50 (22%) (e)	6/49 (12%)	11/50 (22%)
Adjusted Rates (b)	31.2%	17.6%	34.4%
Terminal Rates (c)	10/34 (29%)	6/34 (18%)	11/32 (34%)
Life Table Tests (d)	P=0.490	P=0.139N	P=0.528
Incidental Tumor Tests (d)	P=0.464	P=0.133N	P=0.480
Cochran-Armitage Trend Test (d)	P=0.551		
Fisher Exact Tests		P=0.154N	P=0.595
Pituitary: Adenoma			
Overall Rates (a)	10/50 (20%)	9/48 (19%)	8/49 (16%)
Adjusted Rates (b)	26.3%	24.2%	21.7%
Terminal Rates (c)	7/34 (21%)	6/33 (18%)	4/31 (13%)
Life Table Tests (d)	P=0.446N	P=0.515N	P=0.494N
Incidental Tumor Tests (d)	P=0.475N	P=0.582N	P=0.497N
Cochran-Armitage Trend Test (d)	P=0.366N		
Fisher Exact Tests		P=0.540N	P=0.416N
Pituitary: Carcinoma			
Overall Rates (a)	3/50 (6%)	2/48 (4%)	1/49 (2%)
Adjusted Rates (b)	7.4%	4.2%	3.2%
Terminal Rates (c)	1/34 (3%)	0/33 (0%)	1/31 (3%)
Life Table Tests (d)	P=0.265N	P=0.503N	P=0.355N
Incidental Tumor Tests (d)	P=0.175N	P=0.609	P=0.321N
Cochran-Armitage Trend Test (d)	P=0.229N		
Fisher Exact Tests		P=0.520N	P=0.316N
Pituitary: Adenoma or Carcinoma			
Overall Rates (a)	13/50 (26%)	11/48 (23%)	9/49 (18%)
Adjusted Rates (b)	32.2%	27.4%	24.6%
Terminal Rates (c)	8/34 (24%)	6/33 (18%)	5/31 (16%)
Life Table Tests (d)	P=0.302N	P=0.428N	P=0.342N
Incidental Tumor Tests (d)	P=0.259N	P=0.569	P=0.316N
Cochran-Armitage Trend Test (d)	P=0.215N		
Fisher Exact Tests		P=0.453N	P=0.251N
Adrenal: Pheochromocytoma			
Overall Rates (a)	20/49 (41%)	13/50 (26%)	10/50 (20%)
Adjusted Rates (b)	52.2%	37.1%	30.2%
Terminal Rates (c)	15/33 (45%)	12/34 (35%)	9/32 (28%)
Life Table Tests (d)	P=0.024N	P=0.090N	P=0.038N
Incidental Tumor Tests (d)	P=0.039N	P=0.056N	P=0.084N
Cochran-Armitage Trend Test (d)	P=0.015N		
Fisher Exact Tests		P=0.088N	P=0.021N
Adrenal: Pheochromocytoma or Pheochromocytoma, Malignant			
Overall Rates (a)	20/49 (41%)	13/50 (26%)	11/50 (22%)
Adjusted Rates (b)	52.2%	37.1%	33.2%
Terminal Rates (c)	15/33 (45%)	12/34 (35%)	10/32 (31%)
Life Table Tests (d)	P=0.040N	P=0.090N	P=0.062N
Incidental Tumor Tests (d)	P=0.064N	P=0.056N	P=0.129N
Cochran-Armitage Trend Test (d)	P=0.026N		
Fisher Exact Tests		P=0.088N	P=0.036N

TABLE E1. ANALYSIS OF PRIMARY TUMORS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	250 mg/kg	500 mg/kg
Thyroid: Follicular Cell Adenoma or Carcinoma			
Overall Rates (a)	2/49 (4%)	3/49 (6%)	2/50 (4%)
Adjusted Rates (b)	5.9%	8.8%	6.0%
Terminal Rates (c)	2/34 (6%)	3/34 (9%)	1/32 (3%)
Life Table Tests (d)	P=0.561	P=0.500	P=0.660
Incidental Tumor Tests (d)	P=0.528	P=0.500	P=0.585
Cochran-Armitage Trend Test (d)	P=0.585N		
Fisher Exact Tests		P=0.500	P=0.684N
Thyroid: C-Cell Adenoma			
Overall Rates (a)	7/49 (14%)	5/49 (10%)	3/50 (6%)
Adjusted Rates (b)	19.8%	13.7%	9.4%
Terminal Rates (c)	6/34 (18%)	4/34 (12%)	3/32 (9%)
Life Table Tests (d)	P=0.146N	P=0.381N	P=0.186N
Incidental Tumor Tests (d)	P=0.156N	P=0.420N	P=0.224N
Cochran-Armitage Trend Test (d)	P=0.115N		
Fisher Exact Tests		P=0.380N	P=0.151N
Thyroid: C-Cell Carcinoma			
Overall Rates (a)	5/49 (10%)	0/49 (0%)	1/50 (2%)
Adjusted Rates (b)	14.1%	0.0%	3.1%
Terminal Rates (c)	4/34 (12%)	0/34 (0%)	1/32 (3%)
Life Table Tests (d)	P=0.043N	P=0.035N	P=0.121N
Incidental Tumor Tests (d)	P=0.047N	P=0.027N	P=0.157N
Cochran-Armitage Trend Test (d)	P=0.036N		
Fisher Exact Tests		P=0.028N	P=0.098N
Thyroid: C-Cell Adenoma or Carcinoma			
Overall Rates (a)	12/49 (24%)	5/49 (10%)	4/50 (8%)
Adjusted Rates (b)	33.1%	13.7%	12.5%
Terminal Rates (c)	10/34 (29%)	4/34 (12%)	4/32 (13%)
Life Table Tests (d)	P=0.021N	P=0.057N	P=0.038N
Incidental Tumor Tests (d)	P=0.025N	P=0.058N	P=0.057N
Cochran-Armitage Trend Test (d)	P=0.014N		
Fisher Exact Tests		P=0.054N	P=0.024N
Pancreatic Islets: Islet Cell Adenoma			
Overall Rates (a)	3/50 (6%)	2/49 (4%)	3/50 (6%)
Adjusted Rates (b)	8.6%	5.9%	8.7%
Terminal Rates (c)	2/34 (6%)	2/34 (6%)	2/32 (6%)
Life Table Tests (d)	P=0.552	P=0.503N	P=0.622
Incidental Tumor Tests (d)	P=0.515	P=0.488N	P=0.555
Cochran-Armitage Trend Test (d)	P=0.588		
Fisher Exact Tests		P=0.510N	P=0.661
Pancreatic Islets: Islet Cell Carcinoma			
Overall Rates (a)	1/50 (2%)	1/49 (2%)	3/50 (6%)
Adjusted Rates (b)	2.6%	2.9%	8.9%
Terminal Rates (c)	0/34 (0%)	1/34 (3%)	2/32 (6%)
Life Table Tests (d)	P=0.178	P=0.754	P=0.272
Incidental Tumor Tests (d)	P=0.152	P=0.746N	P=0.199
Cochran-Armitage Trend Test (d)	P=0.202		
Fisher Exact Tests		P=0.747	P=0.309
Pancreatic Islets: Islet Cell Adenoma or Carcinoma			
Overall Rates (a)	4/50 (8%)	3/49 (6%)	6/50 (12%)
Adjusted Rates (b)	10.9%	8.8%	17.1%
Terminal Rates (c)	2/34 (6%)	3/34 (9%)	4/32 (13%)
Life Table Tests (d)	P=0.256	P=0.506N	P=0.318
Incidental Tumor Tests (d)	P=0.210	P=0.479N	P=0.217
Cochran-Armitage Trend Test (d)	P=0.298		
Fisher Exact Tests		P=0.511N	P=0.370

TABLE E1. ANALYSIS OF PRIMARY TUMORS IN MALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	250 mg/kg	500 mg/kg
Mammary Gland: Fibroadenoma			
Overall Rates (a)	8/50 (16%)	2/50 (4%)	2/50 (4%)
Adjusted Rates (b)	21.0%	5.9%	6.2%
Terminal Rates (c)	6/34 (18%)	2/34 (6%)	2/32 (6%)
Life Table Tests (d)	P=0.027N	P=0.050N	P=0.062N
Incidental Tumor Tests (d)	P=0.018N	P=0.053N	P=0.035N
Cochran-Armitage Trend Test (d)	P=0.021N		
Fisher Exact Tests		P=0.046N	P=0.046N
Testis: Interstitial Cell Tumor			
Overall Rates (a)	46/50 (92%)	42/50 (84%)	42/50 (84%)
Adjusted Rates (b)	97.9%	97.6%	100.0%
Terminal Rates (c)	33/34 (97%)	33/34 (97%)	32/32 (100%)
Life Table Tests (d)	P=0.510N	P=0.278N	P=0.565N
Incidental Tumor Tests (d)	P=0.514	P=0.300N	P=0.350
Cochran-Armitage Trend Test (d)	P=0.152N		
Fisher Exact Tests		P=0.179N	P=0.179N

(a) Number of tumor-bearing animals/number of animals examined at the site
(b) Kaplan-Meier estimated tumor incidence at the end of the study after adjusting for intercurrent mortality
(c) Observed tumor incidence at terminal kill
(d) Beneath the vehicle control incidence are the P values associated with the trend test. Beneath the dosed group incidence are the P values corresponding to pairwise comparisons between that dosed group and the vehicle controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as nonfatal. The Cochran-Armitage and Fisher exact test compare directly the overall incidence rates. A negative trend or lower incidence in a dosed group is indicated by (N).
(e) An acinar cell carcinoma was also present in one animal.

TABLE E2. ANALYSIS OF PRIMARY TUMORS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	Vehicle Control	250 mg/kg	500 mg/kg
Hematopoietic System: Mononuclear Cell Leukemia			
Overall Rates (a)	10/50 (20%)	6/50 (12%)	3/50 (6%)
Adjusted Rates (b)	21.7%	14.0%	7.8%
Terminal Rates (c)	5/39 (13%)	3/38 (8%)	2/34 (6%)
Life Table Tests (d)	P=0.046N	P=0.236N	P=0.065N
Incidental Tumor Tests (d)	P=0.014N	P=0.206N	P=0.018N
Cochran-Armitage Trend Test (d)	P=0.025N		
Fisher Exact Tests		P=0.207N	P=0.036N
Pituitary: Adenoma			
Overall Rates (a)	18/50 (36%)	17/50 (34%)	15/50 (30%)
Adjusted Rates (b)	40.7%	39.9%	40.0%
Terminal Rates (c)	13/39 (33%)	13/38 (34%)	12/34 (35%)
Life Table Tests (d)	P=0.466N	P=0.533N	P=0.511N
Incidental Tumor Tests (d)	P=0.309N	P=0.495N	P=0.380N
Cochran-Armitage Trend Test (d)	P=0.298N		
Fisher Exact Tests		P=0.500N	P=0.336N
Pituitary: Carcinoma			
Overall Rates (a)	4/50 (8%)	1/50 (2%)	2/50 (4%)
Adjusted Rates (b)	9.8%	2.2%	5.1%
Terminal Rates (c)	3/39 (8%)	0/38 (0%)	1/34 (3%)
Life Table Tests (d)	P=0.285N	P=0.191N	P=0.402N
Incidental Tumor Tests (d)	P=0.245N	P=0.213N	P=0.345N
Cochran-Armitage Trend Test (d)	P=0.238N		
Fisher Exact Tests		P=0.181N	P=0.339N
Pituitary: Adenoma or Carcinoma			
Overall Rates (a)	22/50 (44%)	18/50 (36%)	17/50 (34%)
Adjusted Rates (b)	48.7%	41.2%	44.0%
Terminal Rates (c)	16/39 (41%)	13/38 (34%)	13/34 (38%)
Life Table Tests (d)	P=0.341N	P=0.321N	P=0.390N
Incidental Tumor Tests (d)	P=0.187N	P=0.280N	P=0.243N
Cochran-Armitage Trend Test (d)	P=0.177N		
Fisher Exact Tests		P=0.270N	P=0.206N
Adrenal: Cortical Adenoma			
Overall Rates (a)	2/50 (4%)	3/50 (6%)	0/50 (0%)
Adjusted Rates (b)	5.1%	7.9%	0.0%
Terminal Rates (c)	2/39 (5%)	3/38 (8%)	0/34 (0%)
Life Table Tests (d)	P=0.237N	P=0.488	P=0.269N
Incidental Tumor Tests (d)	P=0.237N	P=0.488	P=0.269N
Cochran-Armitage Trend Test (d)	P=0.202N		
Fisher Exact Tests		P=0.500	P=0.247N
Adrenal: Cortical Adenoma or Carcinoma			
Overall Rates (a)	3/50 (6%)	3/50 (6%)	0/50 (0%)
Adjusted Rates (b)	7.7%	7.9%	0.0%
Terminal Rates (c)	3/39 (8%)	3/38 (8%)	0/34 (0%)
Life Table Tests (d)	P=0.125N	P=0.652	P=0.146N
Incidental Tumor Tests (d)	P=0.125N	P=0.652	P=0.146N
Cochran-Armitage Trend Test (d)	P=0.101N		
Fisher Exact Tests		P=0.661	P=0.121N
Adrenal: Pheochromocytoma			
Overall Rates (a)	4/50 (8%)	3/50 (6%)	1/50 (2%)
Adjusted Rates (b)	10.3%	7.3%	2.9%
Terminal Rates (c)	4/39 (10%)	2/38 (5%)	1/34 (3%)
Life Table Tests (d)	P=0.168N	P=0.517N	P=0.222N
Incidental Tumor Tests (d)	P=0.166N	P=0.543N	P=0.222N
Cochran-Armitage Trend Test (d)	P=0.133N		
Fisher Exact Tests		P=0.500N	P=0.181N

TABLE E2. ANALYSIS OF PRIMARY TUMORS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	250 mg/kg	500 mg/kg
Adrenal: Pheochromocytoma or Pheochromocytoma, Malignant			
Overall Rates (a)	4/50 (8%)	3/50 (6%)	2/50 (4%)
Adjusted Rates (b)	10.3%	7.3%	5.1%
Terminal Rates (c)	4/39 (10%)	2/38 (5%)	1/34 (3%)
Life Table Tests (d)	P=0.318N	P=0.517N	P=0.398N
Incidental Tumor Tests (d)	P=0.310N	P=0.543N	P=0.392N
Cochran-Armitage Trend Test (d)	P=0.264N		
Fisher Exact Tests		P=0.500N	P=0.339N
Thyroid: Follicular Cell Adenoma or Carcinoma			
Overall Rates (a)	1/50 (2%)	2/50 (4%)	3/50 (6%)
Adjusted Rates (b)	2.6%	5.3%	7.8%
Terminal Rates (c)	1/39 (3%)	2/38 (5%)	2/34 (6%)
Life Table Tests (d)	P=0.188	P=0.491	P=0.269
Incidental Tumor Tests (d)	P=0.236	P=0.491	P=0.452
Cochran-Armitage Trend Test (d)	P=0.222		
Fisher Exact Tests		P=0.500	P=0.309
Thyroid: C-Cell Adenoma			
Overall Rates (a)	5/50 (10%)	4/50 (8%)	4/50 (8%)
Adjusted Rates (b)	12.0%	10.5%	10.8%
Terminal Rates (c)	3/39 (8%)	4/38 (11%)	3/34 (9%)
Life Table Tests (d)	P=0.507N	P=0.514N	P=0.576N
Incidental Tumor Tests (d)	P=0.470N	P=0.540N	P=0.525N
Cochran-Armitage Trend Test (d)	P=0.429N		
Fisher Exact Tests		P=0.500N	P=0.500N
Thyroid: C-Cell Adenoma or Carcinoma			
Overall Rates (a)	5/50 (10%)	6/50 (12%)	6/50 (12%)
Adjusted Rates (b)	12.0%	15.3%	16.5%
Terminal Rates (c)	3/39 (8%)	5/38 (13%)	5/34 (15%)
Life Table Tests (d)	P=0.350	P=0.485	P=0.414
Incidental Tumor Tests (d)	P=0.407	P=0.462	P=0.460
Cochran-Armitage Trend Test (d)	P=0.437		
Fisher Exact Tests		P=0.500	P=0.500
Pancreatic Islets: Islet Cell Adenoma or Carcinoma			
Overall Rates (a)	0/50 (0%)	4/50 (8%)	1/50 (2%)
Adjusted Rates (b)	0.0%	10.1%	2.1%
Terminal Rates (c)	0/39 (0%)	3/38 (8%)	0/34 (0%)
Life Table Tests (d)	P=0.348	P=0.062	P=0.488
Incidental Tumor Tests (d)	P=0.399	P=0.062	P=0.500
Cochran-Armitage Trend Test (d)	P=0.390		
Fisher Exact Tests		P=0.059	P=0.500
Mammary Gland: Fibroadenoma			
Overall Rates (a)	14/50 (28%)	24/50 (50%)	11/50 (22%)
Adjusted Rates (b)	34.8%	55.5%	30.1%
Terminal Rates (c)	13/39 (33%)	19/38 (50%)	9/34 (26%)
Life Table Tests (d)	P=0.469N	P=0.020	P=0.465N
Incidental Tumor Tests (d)	P=0.389N	P=0.019	P=0.433N
Cochran-Armitage Trend Test (d)	P=0.297N		
Fisher Exact Tests		P=0.032	P=0.323N
Mammary Gland: Cystadenoma or Fibroadenoma			
Overall Rates (a)	14/50 (28%)	25/50 (50%)	11/50 (22%)
Adjusted Rates (b)	34.8%	56.6%	30.1%
Terminal Rates (c)	13/39 (33%)	19/38 (50%)	9/34 (26%)
Life Table Tests (d)	P=0.470N	P=0.029	P=0.465N
Incidental Tumor Tests (d)	P=0.376N	P=0.012	P=0.433N
Cochran-Armitage Trend Test (d)	P=0.298N		
Fisher Exact Tests		P=0.020	P=0.323N

TABLE E2. ANALYSIS OF PRIMARY TUMORS IN FEMALE RATS IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	250 mg/kg	500 mg/kg
Clitoral Gland: Carcinoma			
Overall Rates (a)	1/50 (2%)	4/50 (8%)	2/50 (4%)
Adjusted Rates (b)	2.6%	9.4%	5.9%
Terminal Rates (c)	1/39 (3%)	2/38 (5%)	2/34 (6%)
Life Table Tests (d)	P=0.358	P=0.175	P=0.452
Incidental Tumor Tests (d)	P=0.367	P=0.128	P=0.452
Cochran-Armitage Trend Test (d)	P=0.406		
Fisher Exact Tests		P=0.181	P=0.500
Clitoral Gland: Adenoma or Carcinoma			
Overall Rates (a)	1/50 (2%)	5/50 (10%)	2/50 (4%)
Adjusted Rates (b)	2.6%	11.9%	5.9%
Terminal Rates (c)	1/39 (3%)	3/38 (8%)	2/34 (6%)
Life Table Tests (d)	P=0.359	P=0.101	P=0.452
Incidental Tumor Tests (d)	P=0.367	P=0.071	P=0.452
Cochran-Armitage Trend Test (d)	P=0.412		
Fisher Exact Tests		P=0.102	P=0.500
Uterus: Endometrial Stromal Polyp			
Overall Rates (a)	10/50 (20%)	6/50 (12%)	9/50 (18%)
Adjusted Rates (b)	23.4%	15.8%	22.9%
Terminal Rates (c)	7/39 (18%)	6/38 (16%)	6/34 (18%)
Life Table Tests (d)	P=0.549	P=0.228N	P=0.581
Incidental Tumor Tests (d)	P=0.442N	P=0.238N	P=0.418N
Cochran-Armitage Trend Test (d)	P=0.447N		
Fisher Exact Tests		P=0.207N	P=0.500N
Uterus: Endometrial Stromal Sarcoma			
Overall Rates (a)	3/50 (6%)	0/50 (0%)	0/50 (0%)
Adjusted Rates (b)	7.0%	0.0%	0.0%
Terminal Rates (c)	1/39 (3%)	0/38 (0%)	0/34 (0%)
Life Table Tests (d)	P=0.047N	P=0.130N	P=0.153N
Incidental Tumor Tests (d)	P=0.023N	P=0.117N	P=0.074N
Cochran-Armitage Trend Test (d)	P=0.037N		
Fisher Exact Tests		P=0.121N	P=0.121N

(a) Number of tumor-bearing animals/number of animals examined at the site
(b) Kaplan-Meier estimated tumor incidence at the end of the study after adjusting for intercurrent mortality
(c) Observed tumor incidence at terminal kill
(d) Beneath the vehicle control incidence are the P values associated with the trend test. Beneath the dosed group incidence are the P values corresponding to pairwise comparisons between that dosed group and the vehicle controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as nonfatal. The Cochran-Armitage and Fisher exact test compare directly the overall incidence rates. A negative trend or lower incidence in a dosed group is indicated by (N).

TABLE E3. ANALYSIS OF PRIMARY TUMORS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	Vehicle Control	125 mg/kg	250 mg/kg
Skin: Fibroma or Neurofibroma			
Overall Rates (a)	0/50 (0%)	3/50 (6%)	1/50 (2%)
Adjusted Rates (b)	0.0%	7.3%	3.4%
Terminal Rates (c)	0/30 (0%)	3/41 (7%)	1/29 (3%)
Life Table Tests (d)	P=0.361	P=0.181	P=0.493
Incidental Tumor Tests (d)	P=0.361	P=0.181	P=0.493
Cochran-Armitage Trend Test (d)	P=0.378		
Fisher Exact Tests		P=0.121	P=0.500
Lung: Alveolar/Bronchiolar Adenoma			
Overall Rates (a)	6/50 (12%)	7/49 (14%)	7/50 (14%)
Adjusted Rates (b)	17.2%	17.1%	18.4%
Terminal Rates (c)	4/30 (13%)	7/41 (17%)	3/29 (10%)
Life Table Tests (d)	P=0.431	P=0.526N	P=0.497
Incidental Tumor Tests (d)	P=0.323	P=0.618	P=0.351
Cochran-Armitage Trend Test (d)	P=0.442		
Fisher Exact Tests		P=0.484	P=0.500
Lung: Alveolar/Bronchiolar Carcinoma			
Overall Rates (a)	6/50 (12%)	7/49 (14%)	7/50 (14%)
Adjusted Rates (b)	19.3%	17.1%	20.5%
Terminal Rates (c)	5/30 (17%)	7/41 (17%)	4/29 (14%)
Life Table Tests (d)	P=0.426	P=0.501N	P=0.492
Incidental Tumor Tests (d)	P=0.451	P=0.599N	P=0.516
Cochran-Armitage Trend Test (d)	P=0.442		
Fisher Exact Tests		P=0.484	P=0.500
Lung: Alveolar/Bronchiolar Adenoma or Carcinoma			
Overall Rates (a)	11/50 (22%)	13/49 (27%)	13/50 (26%)
Adjusted Rates (b)	32.1%	31.7%	33.5%
Terminal Rates (c)	8/30 (27%)	13/41 (32%)	6/29 (21%)
Life Table Tests (d)	P=0.348	P=0.456N	P=0.408
Incidental Tumor Tests (d)	P=0.287	P=0.584	P=0.313
Cochran-Armitage Trend Test (d)	P=0.364		
Fisher Exact Tests		P=0.385	P=0.408
Hematopoietic System: Malignant Lymphoma, Lymphocytic Type			
Overall Rates (a)	0/50 (0%)	3/50 (6%)	1/50 (2%)
Adjusted Rates (b)	0.0%	6.4%	2.9%
Terminal Rates (c)	0/30 (0%)	1/41 (2%)	0/29 (0%)
Life Table Tests (d)	P=0.379	P=0.155	P=0.500
Incidental Tumor Tests (d)	P=0.335	P=0.060	P=0.536
Cochran-Armitage Trend Test (d)	P=0.378		
Fisher Exact Tests		P=0.121	P=0.500
Hematopoietic System: Malignant Lymphoma, Mixed Type			
Overall Rates (a)	6/50 (12%)	5/50 (10%)	3/50 (6%)
Adjusted Rates (b)	15.5%	11.9%	9.1%
Terminal Rates (c)	2/30 (7%)	4/41 (10%)	2/29 (7%)
Life Table Tests (d)	P=0.196N	P=0.352N	P=0.252N
Incidental Tumor Tests (d)	P=0.212N	P=0.619N	P=0.271N
Cochran-Armitage Trend Test (d)	P=0.195N		
Fisher Exact Tests		P=0.500N	P=0.244N
Hematopoietic System: Lymphoma, All Malignant			
Overall Rates (a)	7/50 (14%)	10/50 (20%)	6/50 (12%)
Adjusted Rates (b)	17.6%	21.9%	18.2%
Terminal Rates (c)	2/30 (7%)	6/41 (15%)	4/29 (14%)
Life Table Tests (d)	P=0.451N	P=0.484	P=0.504N
Incidental Tumor Tests (d)	P=0.475N	P=0.129	P=0.516N
Cochran-Armitage Trend Test (d)	P=0.445N		
Fisher Exact Tests		P=0.298	P=0.500N

TABLE E3. ANALYSIS OF PRIMARY TUMORS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	125 mg/kg	250 mg/kg
Hematopoietic System: Lymphoma or Leukemia			
Overall Rates (a)	7/50 (14%)	11/50 (22%)	6/50 (12%)
Adjusted Rates (b)	17.6%	24.2%	18.2%
Terminal Rates (c)	2/30 (7%)	7/41 (17%)	4/29 (14%)
Life Table Tests (d)	P=0.453N	P=0.403	P=0.504N
Incidental Tumor Tests (d)	P=0.476N	P=0.092	P=0.516N
Cochran-Armitage Trend Test (d)	P=0.446N		
Fisher Exact Tests		P=0.218	P=0.500N
Circulatory System: Hemangiosarcoma			
Overall Rates (a)	7/50 (14%)	2/50 (4%)	3/50 (6%)
Adjusted Rates (b)	19.1%	4.9%	8.8%
Terminal Rates (c)	3/30 (10%)	2/41 (5%)	0/29 (0%)
Life Table Tests (d)	P=0.097N	P=0.041N	P=0.171N
Incidental Tumor Tests (d)	P=0.092N	P=0.124N	P=0.137N
Cochran-Armitage Trend Test (d)	P=0.099N		
Fisher Exact Tests		P=0.080N	P=0.159N
Liver: Hepatocellular Adenoma			
Overall Rates (a)	11/50 (22%)	6/50 (12%)	16/50 (32%)
Adjusted Rates (b)	33.5%	13.9%	49.4%
Terminal Rates (c)	9/30 (30%)	4/41 (10%)	13/29 (45%)
Life Table Tests (d)	P=0.118	P=0.048N	P=0.162
Incidental Tumor Tests (d)	P=0.140	P=0.100N	P=0.174
Cochran-Armitage Trend Test (d)	P=0.139		
Fisher Exact Tests		P=0.144N	P=0.184
Liver: Hepatocellular Carcinoma			
Overall Rates (a)	17/50 (34%)	9/50 (18%)	21/50 (42%)
Adjusted Rates (b)	40.3%	20.6%	50.2%
Terminal Rates (c)	7/30 (23%)	7/41 (17%)	10/29 (34%)
Life Table Tests (d)	P=0.240	P=0.020N	P=0.298
Incidental Tumor Tests (d)	P=0.160	P=0.112N	P=0.192
Cochran-Armitage Trend Test (d)	P=0.225		
Fisher Exact Tests		P=0.055N	P=0.268
Liver: Hepatocellular Adenoma or Carcinoma			
Overall Rates (a)	25/50 (50%)	15/50 (30%)	35/50 (70%)
Adjusted Rates (b)	59.5%	33.1%	82.7%
Terminal Rates (c)	14/30 (47%)	11/41 (27%)	22/29 (76%)
Life Table Tests (d)	P=0.044	P=0.007N	P=0.066
Incidental Tumor Tests (d)	P=0.017	P=0.050N	P=0.017
Cochran-Armitage Trend Test (d)	P=0.029		
Fisher Exact Tests		P=0.033N	P=0.033
Adrenal Capsule: Adenoma			
Overall Rates (a)	2/50 (4%)	4/50 (8%)	3/50 (6%)
Adjusted Rates (b)	6.7%	9.8%	10.3%
Terminal Rates (c)	2/30 (7%)	4/41 (10%)	3/29 (10%)
Life Table Tests (d)	P=0.395	P=0.488	P=0.484
Incidental Tumor Tests (d)	P=0.395	P=0.488	P=0.484
Cochran-Armitage Trend Test (d)	P=0.417		
Fisher Exact Tests		P=0.339	P=0.500
Thyroid: Follicular Cell Adenoma			
Overall Rates (a)	8/48 (17%)	3/50 (6%)	5/50 (10%)
Adjusted Rates (b)	27.6%	7.3%	15.1%
Terminal Rates (c)	8/29 (28%)	3/41 (7%)	3/29 (10%)
Life Table Tests (d)	P=0.193N	P=0.026N	P=0.273N
Incidental Tumor Tests (d)	P=0.190N	P=0.026N	P=0.262N
Cochran-Armitage Trend Test (d)	P=0.188N		
Fisher Exact Tests		P=0.087N	P=0.250N

TABLE E3. ANALYSIS OF PRIMARY TUMORS IN MALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 (Continued)

	Vehicle Control	125 mg/kg	250 mg/kg
Pancreatic Islets: Islet Cell Adenoma			
Overall Rates (a)	1/50 (2%)	0/50 (0%)	4/50 (8%)
Adjusted Rates (b)	3.3%	0.0%	11.5%
Terminal Rates (c)	1/30 (3%)	0/41 (0%)	1/29 (3%)
Life Table Tests (d)	P=0.078	P=0.438N	P=0.190
Incidental Tumor Tests (d)	P=0.107	P=0.438N	P=0.204
Cochran-Armitage Trend Test (d)	P=0.082		
Fisher Exact Tests		P=0.500N	P=0.181
Harderian Gland: Adenoma			
Overall Rates (a)	2/50 (4%)	0/50 (0%)	3/50 (6%)
Adjusted Rates (b)	6.7%	0.0%	9.1%
Terminal Rates (c)	2/30 (7%)	0/41 (0%)	2/29 (7%)
Life Table Tests (d)	P=0.382	P=0.173N	P=0.494
Incidental Tumor Tests (d)	P=0.388	P=0.173N	P=0.497
Cochran-Armitage Trend Test (d)	P=0.390		
Fisher Exact Tests		P=0.247N	P=0.500
All Sites: Neurofibrosarcoma			
Overall Rates (a)	0/50 (0%)	3/50 (6%)	0/50 (0%)
Adjusted Rates (b)	0.0%	6.7%	0.0%
Terminal Rates (c)	0/30 (0%)	1/41 (2%)	0/29 (0%)
Life Table Tests (d)	P=0.639N	P=0.162	(e)
Incidental Tumor Tests (d)	P=0.616N	P=0.070	(e)
Cochran-Armitage Trend Test (d)	P=0.640		
Fisher Exact Tests		P=0.121	(e)

(a) Number of tumor-bearing animals/number of animals examined at the site
(b) Kaplan-Meier estimated tumor incidence at the end of the study after adjusting for intercurrent mortality
(c) Observed tumor incidence at terminal kill
(d) Beneath the vehicle control incidence are the P values associated with the trend test. Beneath the dosed group incidence are the P values corresponding to pairwise comparisons between that dosed group and the vehicle controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as nonfatal. The Cochran-Armitage and Fisher exact test compare directly the overall incidence rates. A negative trend or lower incidence in a dosed group is indicated by (N).
(e) No P value is reported because no tumors were observed in the 250 mg/kg and vehicle control groups.

TABLE E4. ANALYSIS OF PRIMARY TUMORS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3

	Vehicle Control	125 mg/kg	250 mg/kg
Hematopoietic System: Malignant Lymphoma, Mixed Type			
Overall Rates (a)	3/50 (6%)	2/50 (4%)	6/50 (12%)
Adjusted Rates (b)	23.1%	19.8%	41.5%
Terminal Rates (c)	2/12 (17%)	1/8 (13%)	3/9 (33%)
Life Table Tests (d)	P=0.107	P=0.641N	P=0.160
Incidental Tumor Tests (d)	P=0.135	P=0.511N	P=0.195
Cochran-Armitage Trend Test (d)	P=0.169		
Fisher Exact Tests		P=0.500N	P=0.243
Hematopoietic System: Lymphoma, All Malignant			
Overall Rates (a)	4/50 (8%)	5/50 (10%)	6/50 (12%)
Adjusted Rates (b)	24.6%	27.7%	41.5%
Terminal Rates (c)	2/12 (17%)	1/8 (13%)	3/9 (33%)
Life Table Tests (d)	P=0.223	P=0.428	P=0.271
Incidental Tumor Tests (d)	P=0.299	P=0.539	P=0.362
Cochran-Armitage Trend Test (d)	P=0.309		
Fisher Exact Tests		P=0.500	P=0.370
Liver: Hepatocellular Adenoma			
Overall Rates (a)	4/50 (8%)	1/50 (2%)	0/50 (0%)
Adjusted Rates (b)	29.7%	12.5%	0.0%
Terminal Rates (c)	3/12 (25%)	1/8 (13%)	0/9 (0%)
Life Table Tests (d)	P=0.044N	P=0.285N	P=0.092N
Incidental Tumor Tests (d)	P=0.035N	P=0.235N	P=0.072N
Cochran-Armitage Trend Test (d)	P=0.026N		
Fisher Exact Tests		P=0.181N	P=0.059N
Liver: Hepatocellular Adenoma or Carcinoma			
Overall Rates (a)	4/50 (8%)	1/50 (2%)	2/50 (4%)
Adjusted Rates (b)	29.7%	12.5%	14.1%
Terminal Rates (c)	3/12 (25%)	1/8 (13%)	1/9 (11%)
Life Table Tests (d)	P=0.321N	P=0.285N	P=0.429N
Incidental Tumor Tests (d)	P=0.302N	P=0.235N	P=0.396N
Cochran-Armitage Trend Test (d)	P=0.238N		
Fisher Exact Tests		P=0.181N	P=0.339N
Forestomach: Squamous Cell Papilloma			
Overall Rates (a)	0/50 (0%)	0/50 (0%)	3/48 (6%)
Adjusted Rates (b)	0.0%	0.0%	22.8%
Terminal Rates (c)	0/12 (0%)	0/8 (0%)	1/9 (11%)
Life Table Tests (d)	P=0.030	(e)	P=0.092
Incidental Tumor Tests (d)	P=0.031	(e)	P=0.123
Cochran-Armitage Trend Test (d)	P=0.034		
Fisher Exact Tests		(e)	P=0.114
Pituitary: Adenoma			
Overall Rates (a)	4/47 (9%)	2/45 (4%)	6/43 (14%)
Adjusted Rates (b)	30.8%	18.3%	40.5%
Terminal Rates (c)	3/12 (25%)	1/8 (13%)	2/9 (22%)
Life Table Tests (d)	P=0.203	P=0.488N	P=0.256
Incidental Tumor Tests (d)	P=0.291	P=0.346N	P=0.385
Cochran-Armitage Trend Test (d)	P=0.246		
Fisher Exact Tests		P=0.359N	P=0.314
Thyroid: Follicular Cell Adenoma or Carcinoma			
Overall Rates (a)	2/49 (4%)	1/48 (2%)	3/49 (6%)
Adjusted Rates (b)	14.1%	4.5%	33.3%
Terminal Rates (c)	1/12 (8%)	0/8 (0%)	3/9 (33%)
Life Table Tests (d)	P=0.308	P=0.548N	P=0.386
Incidental Tumor Tests (d)	P=0.339	P=0.420N	P=0.429
Cochran-Armitage Trend Test (d)	P=0.400		
Fisher Exact Tests		P=0.508N	P=0.500

TABLE E4. ANALYSIS OF PRIMARY TUMORS IN FEMALE MICE IN THE TWO-YEAR GAVAGE STUDY OF HC RED NO. 3 *(Continued)*

(a) Number of tumor-bearing animals/number of animals examined at the site
(b) Kaplan-Meier estimated tumor incidence at the end of the study after adjusting for intercurrent mortality
(c) Observed tumor incidence at terminal kill
(d) Beneath the vehicle control incidence are the P values associated with the trend test. Beneath the dosed group incidence are the P values corresponding to pairwise comparisons between that dosed group and the vehicle controls. The life table analysis regards tumors in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The incidental tumor test regards these lesions as nonfatal. The Cochran-Armitage and Fisher exact test compare directly the overall incidence rates. A negative trend or lower incidence in a dosed group is indicated by (N).
(e) No P value is reported because no tumors were observed in the 125 mg/kg and vehicle control groups.

APPENDIX F

HISTORICAL INCIDENCES OF TUMORS IN F344/N RATS AND B6C3F$_1$ MICE RECEIVING CORN OIL BY GAVAGE

TABLE F1. HISTORICAL INCIDENCE OF URINARY BLADDER TRANSITIONAL CELL TUMORS IN F344/N RATS RECEIVING CORN OIL BY GAVAGE (a)

MALE

Historical Incidence at Southern Research Institute	No tumors observed in 299 vehicle controls
Overall Historical Incidence	No tumors observed in 1,092 vehicle controls

FEMALE

Historical Incidence at Southern Research Institute

Study	No. of Animals Examined	No. of Tumors in Vehicle Controls	Diagnosis
Allyl isovalerate	49	1	Transitional cell papilloma
All others	247	0	
TOTAL	296	1 (0.3%)	

Overall Historical Incidence

		1	Papilloma, NOS
		1	Transitional cell papilloma
		1	Transitional cell carcinoma
TOTAL	1,084	3 (0.3%)	

(a) Data as of March 16, 1983. No more than one tumor was observed in any vehicle control group.

TABLE F2. HISTORICAL INCIDENCE OF MAMMARY GLAND TUMORS IN FEMALE F344/N RATS RECEIVING CORN OIL BY GAVAGE (a)

Study	Incidence of Fibroadenoma in Vehicle Controls
Historical Incidence at Southern Research Institute	
HC Red No. 3	14/50
Ethyl acrylate	13/50
Benzyl acetate	16/50
Allyl isovalerate	17/50
Allyl isothiocyanate	8/50
Geranyl acetate	12/50
TOTAL	80/300 (26.7%)
SD (b)	6.41%
Range (c)	
High	17/50
Low	8/50
Overall Historical Incidence	
TOTAL	269/1,147 (23.5%)
SD (b)	9.38%
Range (c)	
High	18/50
Low	1/48

(a) Data as of March 16, 1983, for studies of at least 104 weeks
(b) Standard deviation
(c) Range and SD are presented for groups of 35 or more animals.

TABLE F3. HISTORICAL INCIDENCE OF STOMACH TUMORS IN FEMALE B6C3F$_1$ MICE RECEIVING CORN OIL BY GAVAGE (a)

Study	No. of Animals Examined	No. of Tumors in Vehicle Controls	Site	Diagnosis
Incidence at Southern Research Institute				
HC Red No. 3	50	0		
Ethyl acrylate	50	1	Forestomach	Squamous cell papilloma
Benzyl acetate	50	0		
Allyl isovalerate	50	1	Gastric mucosa	Squamous cell papilloma
		1	Gastric mucosa	Adenoma, NOS
Allyl isothiocyanate	47	0		
Geranyl acetate	50	1	Gastric mucosa	Adenomatous polyp, NOS
TOTAL	297	4 (1.3%)		
Overall Historical Incidence (b)				
	1,077	2	Stomach, NOS	Squamous cell papilloma
		1	Stomach, NOS	Adenocarcinoma, NOS
		1	Gastric mucosa	Squamous cell papilloma
		1	Gastric mucosa	Adenoma, NOS
		1	Gastric mucosa	Adenomatous polyp, NOS
		1	Forestomach	Squamous cell papilloma
TOTAL		7 (0.6%)		

(a) Data as of March 16, 1983, for studies of at least 104 weeks
(b) No more than two tumors were observed in any vehicle control group.

TABLE F4. HISTORICAL INCIDENCE OF LIVER TUMORS IN MALE B6C3F$_1$ MICE RECEIVING CORN OIL BY GAVAGE (a)

Study	Incidence in Vehicle Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence at Southern Research Institute			
HC Red No. 3	11/50	17/50	25/50
Ethyl acrylate	6/49	12/49	17/49
Benzyl acetate	0/50	10/50	10/50
Allyl isovalerate	7/50	18/50	23/50
Allyl isothiocyanate	9/49	13/49	21/49
Geranyl acetate	3/50	11/50	13/50
TOTAL	36/298 (12.1%)	81/298 (27.2%)	109/298 (36.6%)
SD (b)	8.06%	6.49%	11.82%
Range (c)			
High	11/50	18/50	25/50
Low	0/50	10/50	10/50
Overall Historical Incidence			
TOTAL	133/1,084 (12.3%)	(d) 222/1,084 (20.5%)	340/1,084 (31.4%)
SD (b)	6.7%	7.9%	10.3%
Range (c)			
High	13/50	18/50	25/50
Low	0/50	4/50	5/50

(a) Data as of March 16, 1983, for studies of at least 104 weeks
(b) Standard deviation
(c) Range and SD are presented for groups of 35 or more animals.
(d) One hepatoblastoma was also observed.

APPENDIX G

CHEMICAL CHARACTERIZATION

OF HC RED NO. 3

I. Identity and Purity Determinations Performed by the Analytical Chemistry Laboratory

A. Lot No. 5890377

1. Physical Properties

a. Appearance: Fine dark maroon crystals

b. Melting Point:

Determined	Literature Values
126.8°-128°C (visual melting point, capillary)	124°-126°C (Clairol Research Labs, personal communication)

2. Spectral Data

a. Infrared

	Determined	Literature Values
(1) Instrument:	Beckman IR-12	
(2) Phase:	1% Potassium bromide pellet	
(3) Results:	See Figure 6	Consistent with that expected for the structure and with the spectra obtained from Clairol Research Labs

b. Ultraviolet/Visible

	Determined		Literature Values	
(1) Instrument:	Cary 118			
(2) Solvent:	Water		Water	
(3) Results:	λ_{max} (nm)	$\varepsilon \times 10^{-3}$	λ_{max} (nm)	$\varepsilon \times 10^{-3}$
	506	4.76 ± 0.04 (δ)	500	
	298 (sh)	4.95 ± 0.15 (δ)	Purified	5.127
	245	19.0 ± 0.4 (δ)	Commercial	4.831
			(Clairol Research Labs)	

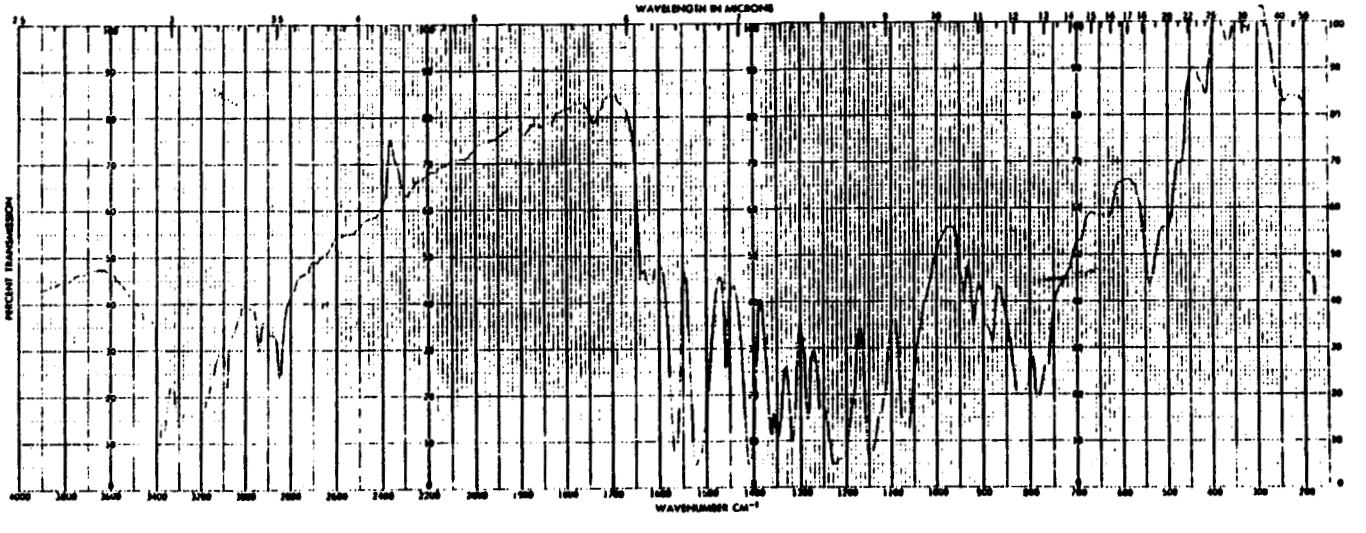

FIGURE 6. INFRARED ABSORPTION SPECTRUM OF HC RED NO. 3 (LOT NO. 5890377)

c. Nuclear Magnetic Resonance

	Determined	Literature Values
(1) Instrument:	Varian EM-360 A	
(2) Solvent:	Deuterated acetone with internal tetramethylsilane	
(3) Assignments:	See Figure 7	No literature reference found. Spectrum is consistent with that expected for structure and with spectra obtained from the manufacturer, except the manufacturer's run in dimethyl sulfoxide-d_6 indicates a different pattern for the exchangeable protons, and the splitting pattern of the (b) proton is obscured.
(4) Chemical Shift (δ):	a t, 3.47 ppm b t, 3.84 ppm c d, 6.87 ppm d d of d, 7.09 ppm e d, 7.42 ppm f s, 4.08 ppm g s, 4.43 ppm h s, 7.94 ppm i s, (impurity) 2.89 ppm j s, (impurity) 3.30 ppm	
(5) Coupling Constant:	J_{ab} = 5 Hz J_{cd} = 9 Hz J_{de} = 2 Hz	
(6) Integration Ratios:	a+j 2.01 b 1.94 c+d 2.08 e 0.97 f+g 2.76 h 1.04 i (impurity) 0.37 j impurity, integrated with (a)	

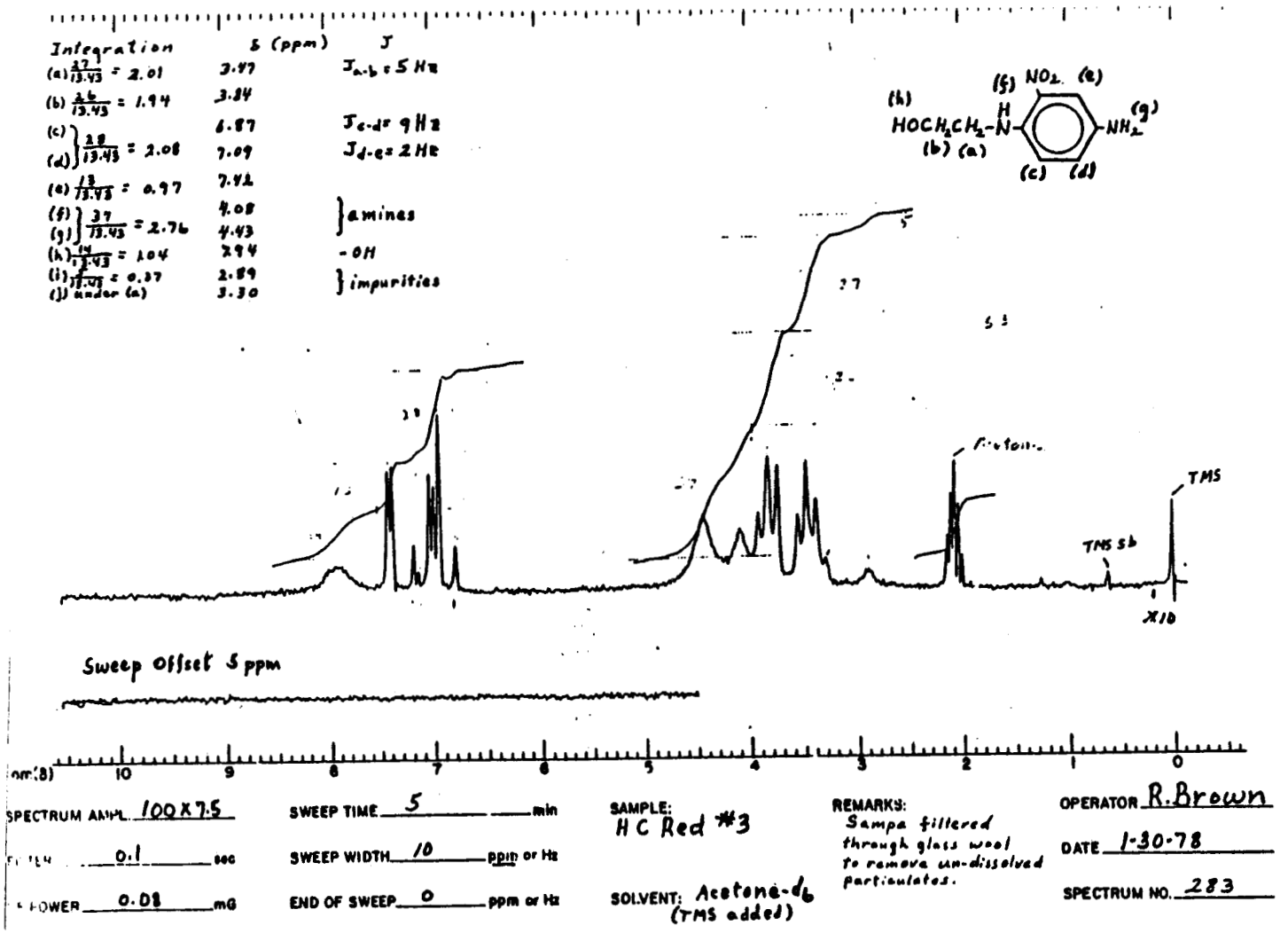

FIGURE 7. NUCLEAR MAGNETIC RESONANCE SPECTRUM OF HC RED NO. 3 (LOT NO. 5890377)

3. Titration: Titration of one amine function with perchloric acid: 98.1% ± 0.7(δ)%

4. Water Analysis (Karl Fischer): 0.22% ± 0.03(δ)%

5. Elemental Analysis

Element	C	H	N
Theory (T)	48.73	5.62	21.31
Determined (D)	48.73	5.55	21.14
	48.90	5.69	21.23
Percent D/T	100.17	100.00	99.41

6. Chromatographic Analysis

a. Thin-Layer Chromatography

(1) Plates: Silica Gel F-60
(2) Reference Standard: 2,6-diaminotoluene
(3) Amount Spotted: 50, 100, and 300 µg, 10 mg/ml in methanol
(4) Visualization: Ultraviolet light (254 and 366 nm); furfural:glacial acetic acid (10 drops:10 ml) (Feigl, 1966)

Spot	R_f	R_{st}

System 1: Chloroform:methanol (78:22)

Trace	0.76	1.05
Minor	0.71	0.98
Major	0.62	0.86
Trace	Origin	Origin

System 2: Ethyl acetate:ethanol (90:10)

Trace	0.60	1.04
Major	0.54	0.94
Minor	0.45	0.78
Trace	0.14	0.24
Trace	Origin	Origin

b. High-Performance Liquid Chromatography

(1) Instrument: Waters Programmable Component System
(2) Column: µBondapak C_{18}, 300 mm × 4 mm, ID
(3) Detection: Ultraviolet, 254 nm
(4) Flow Rate: 1.0 ml/min
(5) Sample Injected: 10 µl of 0.56 mg/ml in methanol
(6) Solvent Program: Acetonitrile:water (25:75), isocratic

(7) Results

Peak No.	Retention Time (min)	Retention Time Relative to Major Peak	Area (percent of major peak)
1	3.5	0.61	0.03
2	4.2	0.74	0.02
3	4.7	0.82	1.2
4	5.7	1.00	100
5	8.9	1.56	0.03
6	15.2	2.67	0.11
7	16.3	2.86	0.03
8	45.0	7.89	1.2

7. Conclusions: Results of elemental analysis for carbon, hydrogen, and nitrogen were in agreement with the theoretical values. Titration of one amine function with perchloric acid indicated a purity of 98.1% ± 0.7(δ)%. High-performance liquid chromatography indicated seven impurities. Two of these each had areas of 1.2% that of the major peak area. The areas of the other five impurities totaled 0.2% that of the major peak area. Thin-layer chromatography with one system indicated one minor and two trace impurities. A second system indicated one minor and three trace impurities. The infrared, ultraviolet/visible, and nuclear magnetic resonance spectra were consistent with the structure and with the spectra submitted by the manufacturer. However, the ratio of ε values at the visible maximum of 500-506 nm indicates this lot to be 93% of the purified dye value, 99% of a typical commercial lot value. The nuclear magnetic resonance spectra also contained two broad impurity peaks.

B. Lot No. C080480

1. Physical Properties

Appearance:	Fine, dark red crystals

2. Spectral Data

a. Infrared	**Determined**	**Literature Values**
(1) Instrument:	Perkin-Elmer 283	
(2) Phase:	1% in Potassium bromide pellet	
(3) Results:	See Figure 8	Consistent with spectrum from Clairol Research Labs and with that expected for the structure

b. Ultraviolet/Visible	**Determined**	**Literature Values**
(1) Instrument:	Cary 118	
(2) Solvent:	Water	
(3) Results:	λ_{max} (nm) / $\varepsilon \times 10^{-3}$	
	506 — 4.68 ± 0.05 (δ)	Consistent with data supplied by the manufacturer
	298 (sh) — 4.77 ± 0.04 (δ)	
	246 — 18.63 ± 0.05 (δ)	

c. Nuclear Magnetic Resonance	**Determined**	**Literature Values**
(1) Instrument:	Varian EM-360A	
(2) Solvent:	Deuterated acetone with internal tetramethylsilane	
(3) Assignments:	See Figure 9	No literature reference found. Spectrum is consistent with that expected for structure and with spectra obtained from the manufacturer.

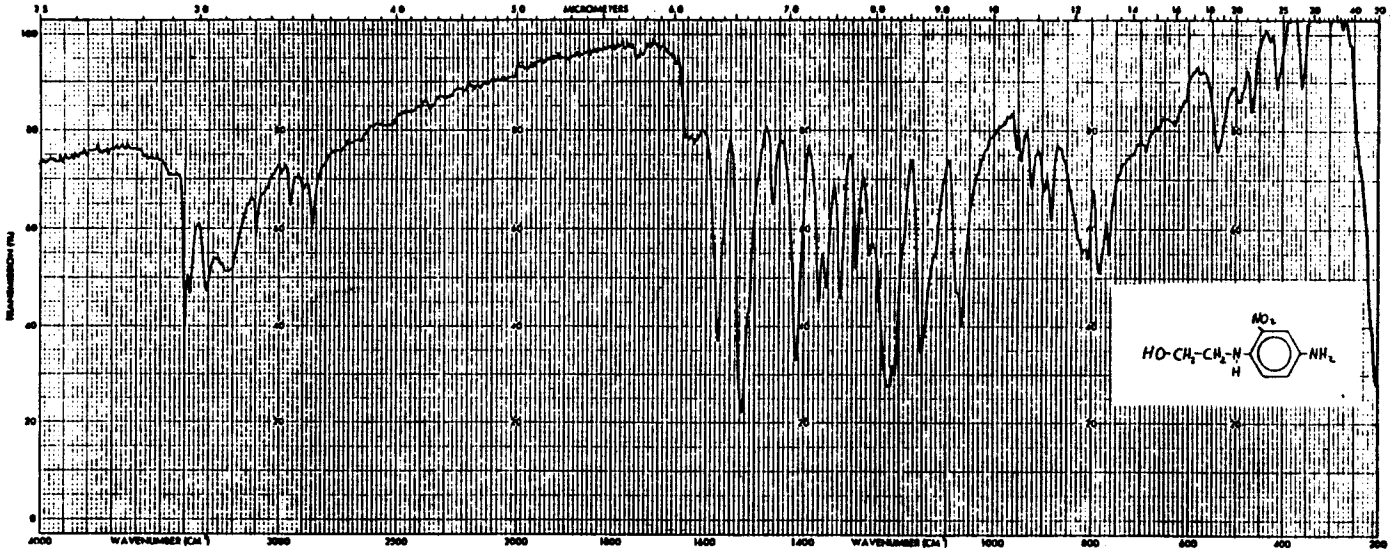

FIGURE 8. INFRARED ABSORPTION SPECTRUM OF HC RED NO. 3 (LOT NO. C080480)

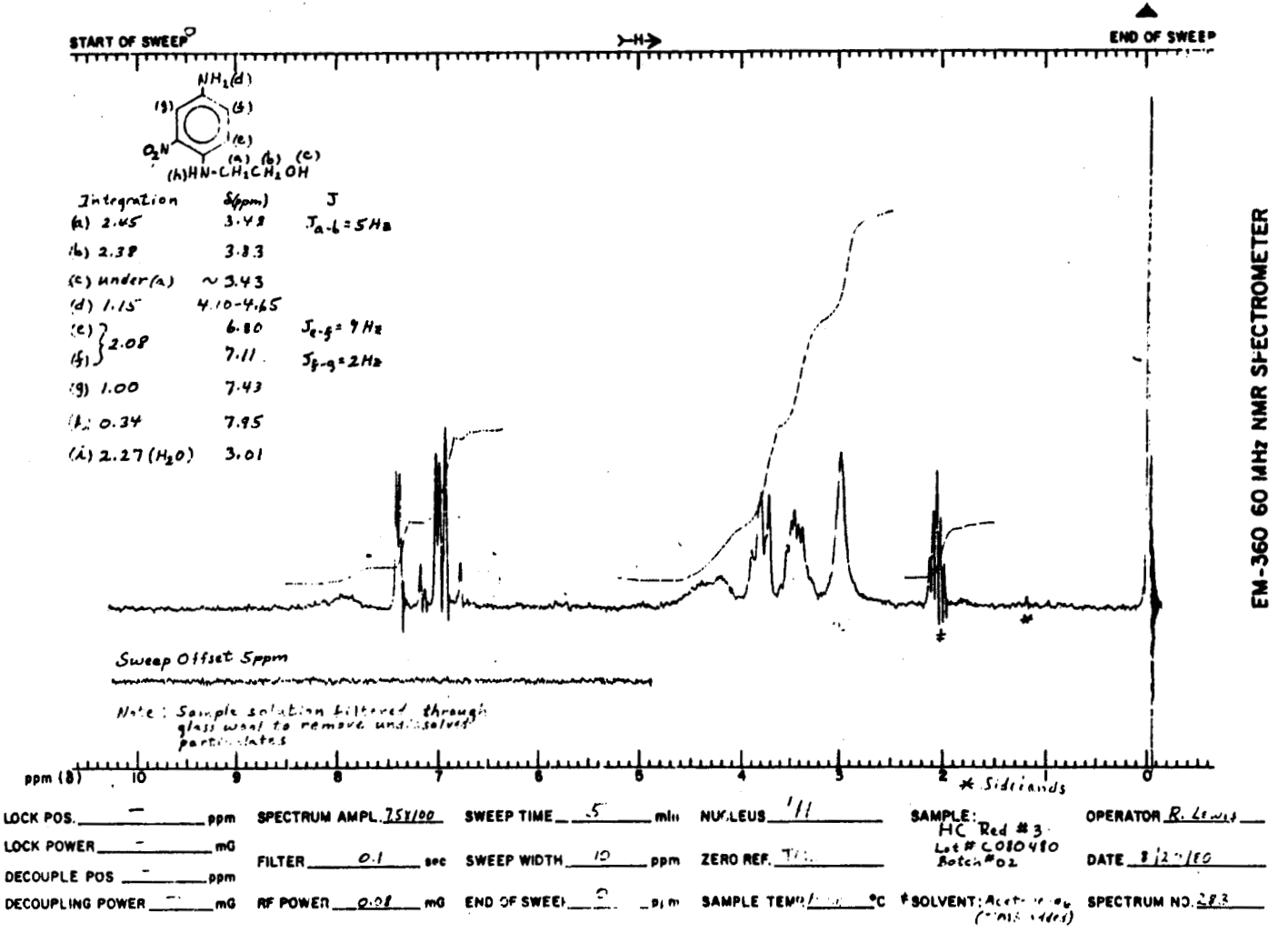

FIGURE 9. NUCLEAR MAGNETIC RESONANCE SPECTRUM OF HC RED NO. 3 (LOT NO. C080480)

(4) Chemical Shift (δ):

a	m,	3.48 ppm
b	m,	3.83 ppm
c*		3.43 ppm
d*	4.10-4.65 ppm	
e	d,	6.8 ppm
f	d of d,	7.11 ppm
g	d,	7.43 ppm
h*	broad s,	7.95 ppm
i*	H_2O,	3.01 ppm

*exchangeable protons

(5) Coupling Constant:

J_{ab} = 5 Hz
J_{ef} = 9 Hz
J_{fg} = 2 Hz

(6) Integration Ratios:

a	2.45
b	2.38
c	under (a)
d	1.15
e, f	2.08
g	1.00
h	0.34
i	2.27 (H_2O)

3. Titration: Titration of one amine function with 0.1N perchloric acid monitored potentiometrically: 97.0% ± 0.5(δ)%

4. Water Analysis (Karl Fischer): 0.21% ± 0.02(δ)%

5. Elemental Analysis

Element	C	H	N
Theory (T)	48.73	5.62	21.31
Determined (D)	48.76	5.58	21.16
	48.93	5.71	21.26
Percent D/T	100.24	100.44	99.53

6. Chromatographic Analysis

a. Thin-Layer Chromatography

(1) Plates: Silica Gel 60, F-254
(2) Reference Standard: 2,6-Diaminotoluene, 1 µl of a 10 mg/ml solution in methanol
(3) Amount Spotted: 1, 10, and 30 µl of a solution (10 mg/ml) in methanol
(4) Visualization: Ultraviolet light (254 nm and 366 nm), visible light; furfural:glacial acetic acid (1 drop:1 ml)

Spot	R_f	R_{st}
System 1: Chloroform:methanol (78:22)		
Minor	0.58	1.07
Major	0.42	0.78
Minor	Origin	
System 2: Ethyl acetate:ethanol (95%) (90:10)		
Minor	0.46	1.09
Major	0.40	0.94
Trace	0.32	0.75
Slight Trace	0.14	0.34
Minor	Origin	

b. High-Performance Liquid Chromatography

(1) Instrument System:
Pump(s): Waters 6000A
Programmer: Waters 660
Detector: Waters 440
Injector: Waters U6K
(2) Column: μBondapak C_{18}, 300 mm × 3.9 mm ID
(3) Detection: Ultraviolet, 254 nm
(4) Guard Column: Whatman CO:Pell ODS 72 mm × 2.3 mm ID
(5) Solvent System: 85% Water:15% acetonitrile
(6) Flow Rate: 1.0 ml/min
(7) Samples Injected: 20 μl of a solution (0.1 mg/ml) of HC Red No. 3 in methanol, filtered
(8) Results

Peak No.	Retention Time (min)	Retention Time Relative to Major Peak	Area (percent of major peak)
1	6.0	0.82	0.41
2	7.3	1.00	100
3	61.3	8.40	2.8

No additional impurities greater than 0.2% of the major peak area were observed with solvent systems of 100%, 80%, 60%, 40%, 20%, 18%, and 16% acetonitrile. At 60% acetonitrile, an impurity was observed eluting after the major peak with an area less than 0.2% that of the major peak area.

The chromatographic profiles indicated the two observed impurities were common to both lots of HC Red No. 3. The impurity eluting before the major peak (retention time = 6 min) was at a concentration approximately one-third as great in lot no. C080480 than in lot no. 5890377, whereas the impurity eluting after the major peak (retention time = 61.3 min) was observed to be nearly three times as large.

7. Conclusion:. The compound was identified as HC Red No. 3 by spectroscopy. Karl Fischer titration indicated 0.21% ± 0.02(δ) % water. Titration of the amine function indicated 97.0% ± 0.5(δ)% (as compared with a value of 98.1% ± 0.7(δ) % for lot no. 5890377. High-performance liquid chromatography indicated two impurities with a combined area of 3.2% of the major peak area. This lot of HC Red No. 3 is comparable in purity with lot no. 5890377.

II. Test Chemical Stability Study Performed by the Analytical Chemistry Laboratory

A. Sample Storage: HC Red No. 3 samples were stored for 2 weeks at – 20°, 5°, 25°, and 65°C.

B. Analytical Method: Duplicate aliquots of each of the above stability samples were accurately weighed into glass-stoppered 100-ml volumetric flasks and diluted to volume with methanol. Then 1.00 ml of this solution was pipetted into a 25-ml volumetric flask and diluted to volume. The solutions then were analyzed by high-performance liquid chromatography.

C. Results

The areas of HC Red No. 3 were compared with the areas of the sample stored at -20° C. The areas were adjusted for the weight of the sample.

Storage Temperature	Percent Purity
-20° C	100.0 ± 6.6 (δ)
5° C	100.7 ± 3.2 (δ)
25° C	101.1 ± 3.0 (δ)
65° C	104.4 ± 1.0 (δ)

D. Conclusion: HC Red No. 3 is stable as the bulk chemical when stored for 2 weeks at temperatures of up to 65°C.

III. Test Chemical Stability at the Testing Laboratory

A. Storage Conditions: The chemical was stored at 22°C.

B. Analytical Method

1. **Purity Determination:** The absorbances of the bulk sample and reference aliquot were determined through the use of a Cary 17 spectrophotometer.

2. **Identity Determination:** The infrared absorption spectra of the samples were obtained as potassium bromide disks by a Perkin-Elmer 621.

C. Results

1. **Purity**

Date of Analysis	Lot No.	Molar Absorptivity (a) Bulk	Molar Absorptivity (a) Reference	Percent of Purity (b)
04/24/78 (c)	54890377	4.00	--	--
07/17/78	54890377	4.26	4.13	103
12/20/78	54890377	4.31	4.58	94
03/05/79	54890377	4.76	4.91	97
07/09/79	54890377	4.39	4.66	94
11/11/79	54890377	4.34	(d) 3.73	(e) 116
03/04/80	54890377	4.43	4.45	99
07/15/80	54890377	4.58	4.49	102
11/05/80 (c)	C080480	4.24	--	--
11/11/80	C080480	4.77	4.49	106
12/07/81	C080480	4.54	4.55	100
	Mean percent purity			
	Lot no. 54890377			98.2 ± 3.9
	Lot no. C080480			103

(a) Molar absorptivity ($\varepsilon \times 10^{-3}$) for the observed λ_{max}, unless otherwise noted at 505 nm
(b) Compared with frozen reference
(c) Initial analysis for lot
(d) λ_{max} at 460 nm
(e) Value not used for delineation of mean

2. **Identity:** All spectra were consistent with the original spectra supplied by the analytical laboratory.

D. Conclusion: No notable degradation occurred during the preliminary or 2-year studies.

APPENDIX H

RECOVERY OF HC RED NO. 3 FROM FORMULATED DIETS

APPENDIX H. RECOVERY

I. Sample Mixing and Storage

A stock solution of HC Red No. 3 in methanol (1.12 mg/ml) was prepared, and 5 ml of this solution was added to individual 5-g samples of Wayne Lab-Blox® Rodent Feed. The methanol was then removed from the samples on a rotary evaporator for 20 minutes at 35° C. Duplicate dried samples were thoroughly mixed with a vortex mixer and were immediately placed in storage for 2 weeks at −20°, 5°, 25°, and 45° C, respectively.

II. Extraction and Analysis

The four pairs (duplicates) of storage samples were equilibrated at room temperature for a minimum of 45 minutes (−20° C samples), up to a maximum of 75 min (45° C samples) before extraction. Each sample was quantitatively transferred to a 200-ml centrifuge bottle and triturated with 50 ml of methanol for 1 minute using a Brinkmann Polytron® high-speed blender. The mixture was then placed in an ultrasonic vibratory bath for 30 seconds and centrifuged for 10 minutes. The supernatant solution was decanted into a 100-ml volumetric flask. The feed residue was mixed with an additional 50 ml of methanol and extracted again as described above. The combined supernatant solutions were brought to volume with additional methanol. A 10-ml aliquot of each extract solution was filtered through a 0.5-μ Millipore filter and analyzed by high-performance liquid chromatography.

Instrument: Waters Programmable Component System
Column: μ-Bondapak C_{18}, 300 × 4 mm, ID
Detector: Ultraviolet, 254 nm
Solvent: Water:acetonitrile (75:25), isocratic
Solvent flow rate: 1.0 ml/min
Retention time of the compound: 5.5 min

III. Quality Control Procedures

Analyses were performed in duplicate for each storage temperature. Room temperature recovery studies were performed in duplicate at the 0.11% concentration for each of three different chemical/feed contact time periods: 0, 1, and 3 hours. Blank (undosed) feed samples were extracted and prepared for analysis in the same manner described above for the test samples. Blanks showed no interference from feed at the retention time of the major component.

IV. Results

A variable time, room-temperature recovery study was performed on HC Red No. 3/feed mixtures in addition to the standard variable temperature, constant time stability study. The timed room temperature results are presented in Table H1; the standard stability test results are in Table H2.

TABLE H1. CONSTANT TEMPERATURE/VARIABLE TIME STUDY OF HC RED NO. 3

Temperature	Contact Time of Chemical on Feed (hours)	Chemical Recovery (percent) (a)
25° C	0	100 ± 4
25° C	1	90 ± 3
25° C	3	79 ± 4

(a) Corrected for the determined value of the −20° C storage sample; 58% ± 4% of the target concentration (0.11% in the diet); assumed to represent 100% stability

TABLE H2. CONSTANT TIME/VARIABLE TEMPERATURE STUDY OF HC RED NO. 3

Temperature	Storage Time (weeks)	Target Concentration (percent, w/w)	Average Concentration (percent w/w) Found in Chemical/Vehicle Mixture Relative to − 20° C Samples (a)
−20° C	2	0.111 ± 0.002	0.111 ± 0.008 (b)
5° C	2	0.111 ± 0.002	0.088 ± 0.006
25° C	2	0.113 ± 0.002	0.041 ± 0.006
45° C	2	0.112 ± 0.002	0.016 ± 0.006

(a) Corrected for the determined value of the −20° C storage sample; 58% ± 4% of the target concentration (0.11% in the diet), assumed to represent 100% stability
(b) Mean ± standard deviation

V. Discussion

Table H1 shows a considerable loss of recoverable chemical from spiked feed mixtures over a period of 3 hours at room temperature. Because of this loss, considered primarily due to physical phenomena rather than chemical transformation, the results reported in Table H2 represent a comparison between the samples stored at the four temperatures, rather than absolute stability data. The entries in Table H2 for the average sample determinations have been corrected for the determined average value of the −20° C storage samples, 58% ± 4% of the target value.

VI. Conclusion

Samples of HC Red No. 3 mixed with stock rodent feed at a concentration of 0.11% and stored for a 2-week period at temperatures of −20°, 5°, 25°, and 45° C, respectively, showed a significant analytical loss of chemical relative to identical chemical/feed samples stored at −20° C. The losses increased with increasing storage temperature and thus are attributed at least in part to chemical instability of HC Red No. 3 at storage temperatures of 5° C and above.

APPENDIX I

STABILITY AND HOMOGENEITY OF HC RED NO. 3 SUSPENDED IN AQUEOUS METHYL CELLULOSE OR CORN OIL

APPENDIX I. STABILITY

I. Seven-Day Room Temperature Stability Studies of HC Red No. 3 in 1% Aqueous Methyl Cellulose

A. Preparation and Storage: HC Red No. 3 (550.0 ± 0.1 mg) was weighed into a 60-ml septum vial. Aqueous methyl cellulose (1%; 50 ml) was added, and the septum vial was reweighed. The mixture was shaken vigorously for 2 minutes and sonicated for 5 minutes, producing a suspension that was visually stable for 10 minutes.

As soon as the suspension was prepared, eight accurately weighed 1.6-g aliquots were removed and transferred to individual 50-ml volumetric flasks. Duplicate aliquots were stored for 1, 3, 5, and 7 days, respectively.

B. Sample Extraction and Analysis: To each 50-ml volumetric flask, methanol was added to the mark. The flask was shaken for 60 seconds and sonicated for 2 minutes. An aliquot (approximately 12 ml) was transferred to a 12-ml centrifuge tube, and the solution was centrifuged for 5 minutes. A 5-ml aliquot was accurately transferred to a second 50-ml volumetric flask and diluted to the mark with methanol. Approximately 2 ml of the above solution was filtered through a 0.5-µ Millipore filter and 18 µl was injected into the high-performance liquid chromatographic system described below.

Instrument: Waters Associates Programmable Component System
Column: µBondapak C_{18}; 300 mm × 4 mm ID
Detector: Ultraviolet, 254 nm
Solvent: Water:Acetonitrile (68:32)
Retention time: 5.6 min

C. Quality Control: Analysis was performed in duplicate. High-performance liquid chromatographic linearity was determined with standard solutions in methanol. Recovery studies at zero time were performed in duplicate at the same concentration level as the test samples.

D. Results and Conclusion

1. Seven-Day Stability Results

Storage Time (days)	Average Percent Chemical Found in Chemical/Vehicle Mixture (a,b)
1	1.12 ± 0.02
3	1.11 ± 0.02
5	1.08 ± 0.02
7	1.12 ± 0.02

(a) 100 ± 1% recovery yield
(b) Target concentration of chemical in methyl cellulose, 1.11%

2. Conclusion: HC Red No. 3 is stable when suspended in an aqueous methyl celullose solution at a 1.1% concentration and stored at room temperature for 7 days.

II. Midwest Research Institute Seven-Day Room Temperature Stability Studies of HC Red No. 3 in Corn Oil

A. Preparation Procedure

1. Sample Preparation and Storage: A suspension of HC Red No. 3 in corn oil was prepared by vigorously shaking for 1 min 553.5 mg of the chemical with 50 ml of corn oil in a 60-ml septum vial. Concentration of HC Red No. 3 in the suspension was 11.07 mg/ml.

A magnetic stirring bar was placed in the vial, and, during the stirring process, 17 aliquots of approximately 1.7 g were transferred to individual 60-ml septum vials and weighed to the nearest 0.1 mg. After being sealed,[a] triplicate vials were set aside for room temperature stability testing after 1, 2, 5, and 7 days' storage. The remaining five vials were used for zero time assays and to confirm homogeneity of the suspension.

2. Extraction and Analysis: Storage samples were extracted by pipetting 50 ml of HPLC-grade methanol into each septum vial and shaking vigorously by hand for 1 minute. About 10 ml of each corn oil suspension was transferred to 12-ml centrifuge tubes and clarified by centrifuging for 5 minutes.

A 5-ml aliquot from each upper methanolic layer was pipetted into individual 50-ml volumetric flasks and diluted to volume with methanol. After being mixed, about 5 ml of solution was filtered through a 0.5 µ Millipore filter and then analyzed for HC Red No. 3 content by the high-performance liquid chromatographic system below:

Instrument: Waters Associates Liquid Chromatograph
Column: µBondapak C_{18}, 300 mm × 4 mm ID
Detector: Ultraviolet, 254 nm
Solvent system: Water:acetonitrile (72:28)
Volume injected: 12 µl
Retention time: 5.6 min

B. Quality Control:

Analyses were carried out by making duplicate injections of duplicate extractions on all samples and recovery determinations. Zero-time recovery studies were conducted with test material at the same concentration as the samples. High-performance liquid chromatographic linearity was determined with standard solutions of HC Red No. 3 at 24.1, 40.2, and 48.2 µg/ml concentrations. Homogeneity of the suspension, determined on five weighings similiar in size as were used for samples, showed a maximum deviation from the mean concentration of only 1.1%.

(a) Vial seals were Microsep F-138 gas chromatography septa with Teflon® film facing, from Canton Biomedical Products, Inc.; the aluminum crimp seals were obtained from Wheaton Scientific Company, Inc.

C. Results and Conclusion

1. Seven-Day Stability Data

Storage Time (Days)	Average Chemical Determined (mg) (a)	Target Milligrams of Chemical	Percent Chemical (b)
0	20.59	20.66	
0	21.31	21.21	
0	20.57	20.69	100 ± 1.1
0	21.08	20.86	
0	20.60	20.72	
1	19.23	20.58	
1	20.23	20.70	95.6 ± 2.1
2	22.06	21.15	
2	21.76	20.85	104.4 ± 0.1
5	19.83	20.57	
5	20.34	20.60	97.6 ± 1.2
7	20.63	20.99	
7	19.73	20.40	97.5 ± 0.8

(a) Results are the average of duplicate injections and were corrected for a zero-time recovery yield of 88.8% ± 0.69 (SD).
(b) Calculated from the assay data corrected for 88.8% recovery yield

2. Conclusion: HC Red No. 3 was found to be stable within the limits of analytical variability of the test in a corn oil gavage suspension at a concentration of 1% after storage for 7 days at room temperature. Excellent homogeneity of suspension was obtained.

II. Studies at the Testing Laboratory

A. Principle of the Method

A weighed quantity of HC Red No. 3 was suspended in a measured volume of corn oil. The resulting suspension was stirred with a magnetic stirring bar and samples were removed for dosing and for analysis. The concentration of HC Red No. 3 in the suspension was determined by spectrophotometric measurement.

B. Procedure

A 1-ml aliquot of the prepared suspension was transferred to a 200-ml volumetric flask. Approximately 150 ml of methanol was added to each flask. These flasks were then shaken, ultrasonicated, and diluted to volume. Aliquots of these solutions were diluted to 50 ml with methanol, and the absorbance of the resulting solutions measured at 492 nm against methanol.

The concentration of HC Red No. 3 in the prepared suspension was determined by comparing the absorbance of the prepared sample with the absorbance of the pure compound. A standard calibration curve of HC Red No. 3 was prepared for this purpose.

C. Results and Conclusions

1. Fourteen-Day Stability Data

Storage Time (days)	Target Milligrams Chemical	Average Milligrams Chemical Found in Sample (individual determinations)	Found/Target (percent)
0	5.00	4.92 (4.85, 5.00)	98.4
7	5.00	4.21 (4.42, 4.00)	84.2
14	5.00	4.74 (4.61, 4.88)	94.8
0	1.25	1.33 (1.30, 1.35)	106.4
7	1.25	1.15 (1.14, 1.16)	92.0
14	1.25	1.19 (1.24, 1.13)	95.2

2. Conclusions

HC Red No. 3 was found to be stable within the limits of the analytical variability of the test in corn oil at the 0.125% and 0.5% concentrations after storage for 14 days at room temperature.

APPENDIX J

METHODS OF ANALYSIS OF DOSE MIXTURES

APPENDIX J. METHODS OF ANALYSIS

The analytical procedures used by the testing and referee laboratories were similar. Both employed a methanolic extraction procedure and spectrophotometric quantitation.

I. Testing Laboratory

Procedure

A weighed quantity of HC Red No. 3 was suspended in a measured volume of corn oil. The resulting suspension was stirred with a magnetic stirring bar, and samples were removed for dosing and for analysis. The samples were shaken on a wrist-action shaker for 1 hour and then stirred for an additional 15 minutes on a magnetic stirrer.

While being stirred, a 1-ml aliquot of the prepared suspension was transferred to a 200-ml volumetric flask. Approximately 150 ml of the methanol was added to each flask. These flasks were then shaken, ultrasonicated, and diluted to volume. Aliquots of these solutions were diluted to 50 ml with methanol, and the absorbance of the resulting solutions was measured at 492 nm against methanol.

The concentration of HC Red No. 3 in the prepared suspension was determined by comparing the absorbance of the prepared sample with the absorbance of the pure compound. A standard calibration curve of HC Red No. 3 was prepared for this purpose.

II. Analytical Chemistry Laboratory

Procedure

A. Preparation of Standard Spiked Corn Oil: One 40-ml aliquot of each of six standard solutions of HC Red No. 3 in methanol was pipetted into individual 60-ml septum vials containing 2 g of undosed corn oil to make spiked corn oil standards bracketing the dose range. One 60-ml septum vial containing 2 g of undosed corn oil was prepared for use as a blank. The spiked corn oil and the corn oil blank were extracted immediately and were analyzed by the procedure below.

B. Preparation of Referee Sample: Three portions (approximately 2 g each) of the referee corn oil suspension sample were transferred to tared 60-ml septum vials and were weighed to the nearest 0.001 g. The samples were extracted immediately and analyzed by the procedure below.

C. Analysis: Forty milliliters of methanol was pipetted into each referee sample, spiked standard, and blank sample vial. After being sealed,[a] the vials were agitated for 60 seconds on a Vortex mixer; then they were vigorously shaken on a Burrell® Model 75 Wrist-Action shaker for 15 minutes. The vials were centrifuged for 5 minutes, and 4-ml aliquots of each upper methanol layer were diluted to 25 ml in methanol. Five-milliliter aliquots were further diluted to 25 ml with methanol. The HC Red No. 3 content of the samples was determined by comparing the absorbance at 493 nm of the solutions and of methanol in 1-cm quartz cells with a Cary 219 spectrophotometer.

D. Quality Assurance: The referee corn oil suspension sample was analyzed in triplicate and the corn oil blank sample was analyzed once. Individually spiked portions of undosed corn oil (six levels) prepared from six independently weighed standards were used to obtain standard curve data.

Results were computed from the linear regression equation obtained for the absorbance of each spiked corn oil sample versus the amount of chemical in the respective spiked corn oil sample. The linearity of the standard curve data was evaluated by the regression equation.

(a) Vial seals were Microsep F-138 gas chromatography septa with Teflon Film facing obtained from Canton Biomedical Products, Inc., Boulder, Colorado 80302; the aluminum crimp seals and vials were available from Wheaton Scientific Company, Inc., Millville, New Jersey.

APPENDIX K

RESULTS OF ANALYSIS OF DOSE MIXTURES

APPENDIX K. RESULTS OF ANALYSIS

I. Thirteen-Week Studies: Dose mixtures were analyzed twice during the 13-week studies. The results ranged from 65% to 113% of the target concentrations.

TABLE K1. CONCENTRATIONS OF HC RED NO.3 IN THE THIRTEEN-WEEK GAVAGE STUDIES

Date Mixed	Concentration (percent w/v) Target	Concentration (percent w/v) Actual	Percent of Target Concentration
04/10/79 (a)			
Rats			
	1.24	1.40	112.9
	2.50	1.95	78.0
	5.00	2.59	51.8
	10.00	3.10	31.0
	20.00	7.60	38.0
Mice			
	0.15	0.10	66.7
	0.31	0.20	65.0
	0.62	0.52	83.9
	1.25	1.18	94.4
	2.50	1.70	68.0
04/24/79			
Rats			
	1.24	1.16	93.6
	2.50	2.14	85.6
	5.00	4.98	99.6
	10.00	(b) 8.55	85.5
	20.00	(b) 17.7	88.5
Mice			
	0.15	(c) 0.16	104
	0.31	0.35	113
	0.62	0.59	94.7
	1.25	1.14	91.2
	2.50	2.47	98.8

(a) The poor results of analysis of the 4/10/79 mix were attributed to poor sampling technique. The method was improved and used for all subsequent dose analyses.
(b) Mean of triplicate samples
(c) Mean of duplicate samples except as noted

II. Two-Year Studies: To estimate the accuracy of dose preparations during the 2-year studies, samples from the dose preparation room were analyzed monthly (Table K2). It is assumed that the number of remixes that were required for the analyzed preparations reflects the total number of mixes that were out of specifications (± 10% of the target concentration).

TABLE K2. CONCENTRATIONS OF HC RED NO. 3 IN THE TWO-YEAR GAVAGE STUDIES (a)

Date Mixed	Determined Concentration for Target Concentration of			
	1.25%(w/v)	2.50%(w/v)	5.00%(w/v)	10.00%(w/v)
12/14/79	1.16		5.50	
01/11/80		2.52		9.80
02/08/80	(b) 1.39		4.81	
02/11/80	(c) 1.28			
03/07/80		2.38		10.4
04/04/80	1.32		4.92	
05/02/80		(d) 2.22		9.42
05/30/80	1.19		4.57	
06/27/80		2.56		10.3
07/25/80	1.30		4.74	
08/22/80		2.48		9.77
09/19/80	1.30		5.15	
10/17/80		2.48		9.33
11/14/80	(e)		(e)	
11/19/80	(c) 1.29		(f) 5.62	
12/12/80		2.72		10.8
01/09/81	(b) 0.97		5.44	
01/15/81	(c) 1.21			
02/06/81		2.37		9.70
03/06/81	1.13		5.04	
04/03/81		2.35	4.88	9.99
05/01/81	1.19		5.14	
05/29/81		(b) 2.82		(b) 12.7
06/05/81		(c) 1.93		
06/26/81	1.25		(b) 4.27	
06/29/81			(c) 4.83	
07/24/81		2.55		9.68
08/21/81	1.22		4.82	
09/18/81		2.28		9.31
10/16/81	1.18		4.86	
Mean (percent w/v)	1.22	2.48	4.93	10.10
Standard deviation	0.109	0.174	0.332	0.935
Coefficient of variation (percent)	8.9	7.0	6.7	9.3
Range (percent w/v)	0.97-1.39	2.22-2.82	4.27-5.50	9.33-12.7
Number of samples	12	12	13	12

(a) The data presented are the results of duplicate analyses.
(b) Out of specifications and not used for dosing. Included in the mean.
(c) Remix. Not included in the mean.
(d) Out of specifications but not remixed. Included in the mean.
(e) Probably sample numbering error. Samples remixed.
(f) Remix out of specifications, used in study; not included in the mean.

APPENDIX K. RESULTS OF ANALYSIS

To confirm the accuracy of dose preparation, aliquots taken from the animal room dosing vials were analyzed by the testing laboratory and Midwest Research Institute for referee analysis. All six samples from the animal room were within 10% of the reported dose preparation room samples (Table K3).

The initial two referee analyses gave poor agreement with the testing laboratory. After what appeared to be a sampling problem was resolved, all subsequent data showed agreement between the laboratories (Table K4).

TABLE K3. RESULTS OF ANALYSIS OF DOSE PREPARATION ROOM SAMPLES AND ANIMAL ROOM SAMPLES IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3 (a)

Date Mixed	Target Concentration (percent, wt/vol)	Determined Concentration of HC Red No. 3 in Corn Oil	
		Dose Preparation Room Samples	Animal Room Samples
07/25/80	1.25	1.30	1.30
	5.00	4.74	4.74
07/24/81	2.50	2.43	2.56
	10.00	9.78	9.68
08/21/81	1.25	1.22	1.13
	5.00	4.82	5.28

(a) Southern Research Institute data

TABLE K4. RESULTS OF REFEREE ANALYSIS OF DOSE MIXTURES OF HC RED NO. 3 IN CORN OIL IN THE TWO-YEAR GAVAGE STUDIES

Date Mixed	Target Concentration (percent, wt/vol)	Determined Concentration	
		Testing Laboratory (a)	Analytical Laboratory (b)
03/07/80	2.5	2.38	1.86
04/04/80	5.0	4.92	3.87
08/22/80	10.0	9.78	9.30
01/09/81	5.0	5.44	5.31
07/24/81	2.5	2.44	2.54
10/16/81	1.25	1.18	1.15

(a) Results of duplicate analysis
(b) Results of triplicate analysis

APPENDIX L

SENTINEL ANIMAL PROGRAM

APPENDIX L. SENTINEL ANIMAL PROGRAM

I. Methods

Rodents used in the Carcinogenesis Program of the National Toxicology Program are produced in optimally clean facilities to eliminate potential pathogens that may affect test results. The Sentinel Animal Program is part of the periodic monitoring of animal health that occurs during the toxicologic evaluation of chemical compounds. Under this program, the disease state of the rodents is monitored via viral serology on sera from extra (sentinel) animals in the test rooms. These animals are untreated, and these animals and the test animals are both subject to identical environmental conditions. The sentinel animals come from the same production source and weanling groups as the animals used for the studies of chemical compounds.

Fifteen $B6C3F_1$ mice and 15 F344/N rats of each sex are selected at the time of randomization and allocation of the animals to the various study groups. Five animals of each designated sentinel group are killed at 6, 12, and 18 months on study. Data from animals surviving 24 months are collected from 5/50 randomly selected control animals of each sex and species. The blood from each animal is collected and clotted, and the serum is separated. The serum is cooled on ice and shipped to Microbiological Associates' Comprehensive Animal Diagnostic Service for determination of the viral antibody titers. The following tests are performed:

	Hemagglutination Inhibition	Complement Fixation	ELISA
Mice	PVM (pneumonia virus of mice) Reo 3 (reovirus type 3) GDVII (Theiler's encephalomyelitis virus) Poly (polyoma virus) MVM (minute virus of mice) Ectro (infectious ectromelia) Sendai	M.Ad. (mouse adenovirus) LCM (lymphocytic choriomeningitis virus) Sendai (24 mo)	MHV (mouse hepatitis virus)
Rats	PVM KRV (Kilham rat virus) H-1 (Toolan's H-1 virus) Sendai	RCV (rat coronavirus) Sendai (24 mo)	

II. Results

Results are presented in Table L1.

TABLE L1. MURINE VIRUS ANTIBODY DETERMINATIONS FOR RATS AND MICE IN THE TWO-YEAR GAVAGE STUDIES OF HC RED NO. 3 (a)

	Interval (months)	No. of Animals	Positive Serologic Reaction for
RATS			
	6	--	None positive
	12	--	None positive
	18	--	None positive
	24	--	None positive
MICE			
	6	--	None positive
	12	--	None positive
	18	--	None positive
	24	2/10	MHV

(a) Blood samples were taken from sentinel animals at approximately 6, 12, and 18 months after the start of dosing and from the vehicle control animals just before they were killed; samples were sent to Microbiological Associates, Inc. (Bethesda, MD) for the Animal Disease Screening Program.

APPENDIX M

GENETIC TOXICOLOGY OF

HC RED NO. 3

TABLE M1. MUTAGENICITY OF HC RED NO. 3 IN *SALMONELLA TYPHIMURIUM*

Strain	Dose (µg/plate)	Revertants/Plate (a,b) −S9	+S9 (rat)	+S9 (hamster)
TA100	0	139 ± 14.8	99 ± 3.5	105 ± 10.7
	33	138 ± 7.9	118 ± 5.8	158 ± 9.0
	100	144 ± 2.9	223 ± 19.1	251 ± 8.4
	333	191 ± 7.8	534 ± 16.6	454 ± 6.6
	1,000	272 ± 5.6	1,030 ± 17.9	967 ± 14.4
	3,333	354 ± 4.8	841 ± 26.6	931 ± 22.7
TA1535	0	34 ± 4.5	12 ± 1.3	11 ± 2.0
	33	34 ± 3.5	12 ± 0.9	11 ± 3.2
	100	33 ± 5.6	12 ± 1.5	15 ± 1.5
	333	29 ± 4.2	16 ± 1.8	13 ± 1.5
	1,000	28 ± 3.9	16 ± 0.9	18 ± 0.9
	3,333	27 ± 3.9	9 ± 1.2	13 ± 2.1
TA97	0	108 ± 9.3	128 ± 4.5	153 ± 3.8
	3.3	--	207 ± 3.0	213 ± 11.7
	10	--	323 ± 13.9	314 ± 1.0
	33	148 ± 4.8	939 ± 4.9	819 ± 26.0
	100	186 ± 11.7	2,171 ± 60.6	1,637 ± 33.2
	333	248 ± 2.3	3,947 ± 159.8	3,565 ± 180.6
	1,000	391 ± 9.5	--	--
	3,333	401 ± 10.4	--	--
TA98	0	26 ± 3.9	36 ± 3.2	37 ± 3.0
	3.3	--	131 ± 0.9	105 ± 10.9
	10	--	326 ± 5.5	300 ± 17.0
	33	45 ± 6.7	1,220 ± 13.4	1,105 ± 23.3
	100	68 ± 5.9	2,764 ± 33.5	2,723 ± 83.1
	333	160 ± 4.2	4,212 ± 67.2	4,366 ± 40.1
	1,000	307 ± 3.9	--	--
	3,333	383 ± 28.0	--	--

(a) The S9 fractions were prepared from the livers of Aroclor 1254-induced male Sprague-Dawley rats and male Syrian hamsters. Cells and test compound or solvent (DMSO) were incubated for 20 min at 37° C in the presence of either S9 or buffer. After the addition of soft agar, the contents of each tube were poured onto minimal medium, and the plates were incubated at 37° C for 48 h (Haworth et al., 1983). The experiment was performed twice, each in triplicate; because the results were similar, data from only one experiment are shown.
(b) Mean ± standard error

APPENDIX N

INGREDIENTS, NUTRIENT COMPOSITION, AND CONTAMINANT LEVELS OF THE NIH 07 DIET

Pelleted Diet: December 1979 to January 1982

(Manufactured by Zeigler Bros., Inc., Gardners, PA)

TABLE N1. INGREDIENTS OF THE NIH 07 DIET (a)

Ingredients (b)	Percent by Weight
Ground #2 yellow shelled corn	24.50
Ground hard winter wheat	23.00
Soybean meal (49% protein)	12.00
Fish meal (60% protein)	10.00
Wheat middlings	10.00
Dried skim milk	5.00
Alfalfa meal (dehydrated, 17% protein)	4.00
Corn gluten meal (60% protein)	3.00
Soy oil	2.50
Brewer's dried yeast	2.00
Dry molasses	1.50
Dicalcium phosphate	1.25
Ground limestone	0.50
Salt	0.50
Premixes (vitamin and mineral)	0.25

(a) NIH, 1978; NCI, 1976
(b) Ingredients should be ground to pass through a U.S. Standard Screen #16 before mixing.

TABLE N2. VITAMINS AND MINERALS IN THE NIH 07 DIET (a)

	Amount	Source
Vitamins		
A	5,500,000 IU	Stabilized vitamin A palmitate or acetate
D_3	4,600,000 IU	D activated animal sterol
d-A-tocopheryl acetate	20,000 IU	
Riboflavin	3.4 g	
Thiamine	10.0 g	Thiamine mononitrate
Niacin	30.0 g	
d-Pantothenic acid	18.0 g	d-Calcium pantothenate
Folic acid	2.2 g	
Pyridoxine	1.7 g	Pyridoxine hydrochloride
B_{12}	4000 µg	
Biotin	140.0 mg	d-Biotin
K_3	2.8 g	Menadione activity
Choline	560.0 g	Choline chloride
Minerals		
Iron	120.0 g	Iron sulfate
Manganese	60.0 g	Manganous oxide
Zinc	16.0 g	Zinc oxide
Copper	4.0 g	Copper sulfate
Iodine	1.4 g	Calcium iodate
Cobalt	0.4 g	Cobalt carbonate

(a) Per ton (2,000 lb) of finished product

TABLE N3. NUTRIENT COMPOSITION OF THE NIH 07 DIET (a)

Nutrient	Mean ± Standard Deviation	Range	Number of Samples
Crude protein (percent by weight)	24.29 ± 0.81	22.7-26.1	24
Crude fat (percent by weight)	4.81 ± 0.38	4.1-5.5	24
Crude fiber (percent by weight)	3.31 ± 0.50	1.4-4.3	24
Ash (percent by weight)	6.76 ± 0.44	5.83-7.43	24
Vitamins			
Vitamin A (IU/kg)	10,192 ± 2,534	6,700-17,000	24
Vitamin D (IU/kg)	6,300		1
A-tocopherol (ppm)	37.6	31.1-44.0	2
Thiamine (ppm) (b)	16.2 ± 4.5	7.4-27.0	24
Riboflavin (ppm)	6.9	6.1-7.4	2
Niacin (ppm)	75	65-85	2
Pantothenic acid (ppm)	30.2	29.8-30.5	2
Pyridoxine (ppm)	7.2	5.6-8.8	2
Folic acid (ppm)	2.1	1.8-2.4	2
Biotin (ppm)	0.24	0.21-0.27	2
Vitamin B_{12} (ppb)	12.8	10.6-15.0	2
Choline (ppm)	3,315	3,200-3,430	2
Minerals			
Calcium (percent)	1.34 ± 0.20	0.81-1.69	24
Phosphorous (percent)	1.01 ± 0.08	0.82-1.10	24
Potassium (percent)	0.809	0.772-0.846	2
Chloride (percent)	0.557	0.479-0.635	2
Sodium (percent)	0.304	0.258-0.349	2
Magnesium (percent)	0.172	0.166-0.177	2
Sulfur (percent)	0.278	0.270-0.285	2
Iron (ppm)	418	409-426	2
Manganese (ppm)	90.8	86.0-95.5	2
Zinc (ppm)	55.1	54.2-56.0	2
Copper (ppm)	12.68	9.65-15.70	2
Iodine (ppm)	2.58	1.52-3.64	2
Chromium (ppm)	1.86	1.79-1.93	2
Cobalt (ppm)	0.57	0.49-0.65	2
Essential Fatty Acids (percent of total diet)			
Linoleic	2.37		1
Linolenic	0.308		1
Arachidonic	0.008		1
Essential Amino Acids (percent of total diet)			
Arginine	1.260	1.21-1.31	2
Cystine	0.395	0.39-0.40	2
Glycine	1.175	1.15-1.20	2
Histidine	0.553	0.530-0.576	2
Isoleucine	0.908	0.881-0.934	2
Leucine	1.905	1.85-1.96	2
Lysine	1.250	1.20-1.30	2
Methionine	0.310	0.306-0.314	2
Phenylalanine	0.967	0.960-0.974	2
Threonine	0.834	0.840 - 0.827	2
Tryptophan	0.175	0.171-0.178	2
Tyrosine	0.587	0.566-0.607	2
Valine	1.085	1.05-1.12	2

(a) One or two of the analyzed feed batches came from diet manufactured in January and/or April 1983.
(b) One batch (7/22/81) was not analyzed for thiamine.

TABLE N4. CONTAMINANT LEVELS OF THE NIH 07 DIET

Contaminant	Mean ± Standard Deviation	Range	Number of Samples
Arsenic (ppm)	0.39 ± 0.23	<0.05-1.06	24
Cadmium (ppm)	0.11 ± 0.07	(a) <0.05-0.40	24
Lead (ppm)	0.91 ± 0.51	0.50-2.65	24
Mercury (ppm)	(b) <0.05		
Selenium (ppm)	0.29 ± 0.09	0.10-0.52	24
Aflatoxins (ppb)	(b)(c) <10		24
Nitrate nitrogen (ppm) (d)	7.00 ± 3.70	(e) <0.1-13.0	24
Nitrite nitrogen (ppm) (d)	1.45 ± 1.02	(e) <0.1-4.0	24
BHA (ppm) (f)	3.83 ± 3.88	(g) <0.2-13.0	24
BHT (ppm) (f)	2.97 ± 1.74	0.8-7.6	24
Aerobic plate count (CFU/g)	48,786 ± 32,701	(h) 5,500-120,000	22
	70,970 ± 81,410	(i) 5,500-320,000	24
Coliform (MPN/g)	39 ± 57	(j) <3-240	20
	270 ± 580	(k) <3-2,400	24
E. Coli (MPN/g)	(l)<3		24
Total nitrosamines (ppb)	7.63 ± 6.67	(m, n) 2.2-24.5	21
	29.77 ± 64.59	(m, o) 2.2-273	24
N-Nitrosodimethylamine (ppb)	5.81 ± 6.30	(m, n) 1.1-20.0	21
	27.79 ± 64.31	(m, o) 1.1-272	24
N-Nitrosopyrrolidine (ppb)	1.44 ± 0.89	0.5-3.5	24
Pesticides (ppm)			
Alpha BHC (p)	(b) <0.01		24
Beta BHC	(b) <0.02		24
Gamma BHC-Lindane	(b) <0.01		24
Delta BHC	(b) <0.01		24
Heptachlor	(b) <0.01		24
Aldrin	(b) <0.01		24
Heptachlor epoxide	(b) <0.01		24
DDE	(b) <0.01		24
DDD	(b) <0.01		24
HCB	(b) <0.01		24
Mirex	(b) <0.01		24
Methoxychlor	(b) <0.05	(q) 0.09 (8/26/81)	24
Dieldrin	(b) <0.01		24
Endrin	(b) <0.01		24
Telodrin	(b) <0.01		24
Chlordane	(b) <0.05		24
Toxaphene	(b) <0.1		24
Estimated PCB's	(b) <0.2		24
Ronnel	(b) <0.01		24
Ethion	(b) <0.02		24
Trithion	(b) <0.05		24
Diazinon	(b) <0.1	(q) 0.2 (4/27/81)	24
Methyl parathion	(b) <0.02		24
Ethyl parathion	(b) <0.02		24
Malathion	0.10 ± 0.07	(r) <0.05 - 0.27	24
Endosulfan I	(b) <0.01		24
Endosulfan II	(b) <0.01		24
Endosulfan sulfate	(b) <0.03		24

TABLE N4. CONTAMINANT LEVELS OF THE NIH 07 DIET (Continued)

(a) Three batches contained more than 0.1 ppm.
(b) All values less than detection limit, which is given in the table as the mean
(c) Detection limit reduced from 10 ppb to 5 ppb after 7/81
(d) Source of contamination--alfalfa, grains, and fish meal
(e) Two batches contained less than 0.1 ppm.
(f) Source of contamination--soy oil and fish meal
(g) Six batches contained less than 0.5 ppm.
(h) Mean, standard deviation (SD), and range excludes two extreme values (300,0000 and 320,000) obtained in batches produced on 12/21/79 and 2/26/80; CFU = colony-forming units.
(i) Mean, SD, and range includes the two extreme values given in (h).
(j) Excludes four very high values in the range of 1,100-2,400 obtained in batches produced on 2/4/80, 2/26/80, 5/29/80, and 12/16/80
(k) Includes the high values listed in (j)
(l) All values were less than 3 MPN/g (MPN = most probable number).
(m) All values were corrected for percent recovery.
(n) Mean, SD, and range excludes three very high values in the range of 115-280 ppb in batches produced on 1/26/81, 2/23/81, and 4/27/81.
(o) Mean, SD, and range includes the very high values given in (n).
(p) BHC is hexachlorocyclohexane or benzene hexachloride.
(q) One observation was above the detection limit. The value and the date it was obtained are given under the range.
(r) Nine batches contained more than 0.05 ppm.

APPENDIX O

DATA AUDIT SUMMARY

APPENDIX O. DATA AUDIT SUMMARY

The experimental data and tables of the NTP Technical Report on the toxicology and carcinogenesis studies of HC Red No. 3 in F344/N rats and B6C3F$_1$ mice were examined for completeness, consistency, and accuracy and for procedures consistent with Good Laboratory Practice guidelines. The audit was conducted by ImmuQuest Laboratories, Inc. The following people were involved in the audit: P.H. Errico, M.A.; C.S. Reese, M.S.; K.M. Witkin, Ph.D.; L.H. Brennecke, D.V.M.; and D.C. Haynes, H.T. The 2-year studies in rats and mice were conducted between November 1979 and December 1981 at Southern Research Institute, Birmingham, Alabama, under a subcontract with Tracor Jitco, Inc.

The full report of the audit is on file at the National Toxicology Program, NIEHS. The audit included a review of the records of the in-life portion of the studies for 10% of the animals, 100% of the analytical chemistry data, and a random 50% sample of the chemical mix calculations. All Individual Animal Data Records were examined for correspondence between necropsy observations and histopathologic findings. All wet tissue bags were counted, and 10% were reviewed for animal identification and the presence of untrimmed lesions. A complete slide-block match for both sexes of both species in the vehicle control and high dose groups was performed.

The audit revealed no major problems with the conduct of the studies or with collection and documentation of the experimental data. It was not possible to confirm the identification of one rat that had a portion of one ear missing, and some discrepancies between gross and microscopic diagnoses in nontarget organs were noted. Minor problems or discrepancies, considered inconsequential for the interpretation of the studies, were not necessarily pursued to final resolution but are identified in the NTP audit report. In conclusion, the data examined during this audit are considered adequate to support the conclusions presented in the Technical Report.

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