NTP TECHNICAL REPORT

ON THE

TOXICOLOGY AND CARCINOGENESIS

STUDIES OF

D&C YELLOW NO. 11

(CAS NO. 8003-22-3)

IN F344/N RATS

(FEED STUDIES)

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

April 1997

NTP TR 463

NIH Publication No. 97-3379

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

FOREWORD

The National Toxicology Program (NTP) is made up of four charter agencies of the U.S. Department of Health and Human Services (DHHS): 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. The NTP coordinates the relevant programs, staff, and resources from these Public Health Service agencies relating to basic and applied research and to biological assay development and validation.

The NTP develops, evaluates, and disseminates scientific information about potentially toxic and hazardous chemicals. This knowledge is used for protecting the health of the American people and for the primary prevention of disease.

The studies described in this Technical Report were performed under the direction of the NIEHS and were conducted in compliance with NTP laboratory health and safety requirements and must meet or exceed all applicable federal, state, and local health and safety regulations. Animal care and use were in accordance with the Public Health Service Policy on Humane Care and Use of Animals. The prechronic and chronic studies were conducted in compliance with Food and Drug Administration (FDA) Good Laboratory Practice Regulations, and all aspects of the chronic studies were subjected to retrospective quality assurance audits before being presented for public review.

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 NTP toxicology and carcinogenesis studies are chosen primarily on the bases of human exposure, level of production, and chemical structure. The interpretive conclusions presented in this Technical Report are based only on the results of these NTP studies. Extrapolation of these results to other species and quantitative risk analyses for humans require wider analyses beyond the purview of these studies. Selection *per se* is not an indicator of a chemical's carcinogenic potential.

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 while supplies last from NTP Central Data Management, NIEHS, P.O. Box 12233, MD E1-02, Research Triangle Park, NC 27709 (919-541-3419). Listings of all published NTP reports and ongoing studies are also available from NTP Central Data Management. The Abstracts and other study information for 2-year studies are also available at the NTP's World Wide Web site: http://ntp-server.niehs.nih.gov.

NTP TECHNICAL REPORT

ON THE

TOXICOLOGY AND CARCINOGENESIS

STUDIES OF

D&C YELLOW NO. 11

(CAS NO. 8003-22-3)

IN F344/N RATS

(FEED STUDIES)

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

April 1997

NTP TR 463

NIH Publication No. 97-3379

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

CONTRIBUTORS

National Toxicology Program
Evaluated and interpreted results and reported findings

W.C. Eastin, Ph.D., Study Scientist
G.A. Boorman, D.V.M., Ph.D.
D.A. Bridge, B.S.
J.R. Bucher, Ph.D.
L.T. Burka, Ph.D.
R.E. Chapin, Ph.D.
J.R. Hailey, D.V.M.
J.K. Haseman, Ph.D.
A. Radovsky, D.V.M., Ph.D.
G.N. Rao, D.V.M., Ph.D.
J.H. Roycroft, Ph.D.
G.S. Travlos, D.V.M.
D.B. Walters, Ph.D.
K.L. Witt, M.S., Oak Ridge Associated Universities

Southern Research Institute
Conducted studies, evaluated pathology findings

J.D. Prejean, Ph.D., Principal Investigator
D.G. Serota, Ph.D., Principal Investigator
J.E. Heath, D.V.M.
R. Hoar, Ph.D., Argus Research Laboratories, Inc.
C. Lindamood, III, Ph.D.

Experimental Pathology Laboratories, Inc.
Provided pathology quality assurance

J.F. Hardisty, D.V.M., Principal Investigator
C.C. Shackelford, D.V.M., M.S., Ph.D.

Dynamac Corporation
Prepared quality assurance audits

S. Brecher, Ph.D., Principal Investigator

Analytical Sciences, Inc.
Provided statistical analyses

R.W. Morris, M.S., Principal Investigator
N.G. Mintz, B.S.
S. Rosenblum, M.S.

NTP Pathology Working Group
Evaluated slides, prepared pathology report on rats (4 April 1995)

M.P. Jokinen, D.V.M., Chairperson
Pathology Associates, Inc.
R.C. Cattley, M.S., V.M.D., Ph.D.
Chemical Industry Institute of Toxicology
M.R. Elwell, D.V.M., Ph.D.
National Toxicology Program
L. Gaboury, M.D., Ph.D., Observer
Universite de Montreal
J.R. Hailey, D.V.M.
National Toxicology Program
R.A. Herbert, D.V.M., Ph.D.
National Toxicology Program
R.R. Maronpot, D.V.M.
National Toxicology Program
A. Radovsky, D.V.M., Ph.D.
National Toxicology Program
C.C. Shackelford, D.V.M., M.S., Ph.D.
Experimental Pathology Laboratories, Inc.

Evaluated slides, prepared pathology report on extended renal evaluation in rats (19 September 1995)

J.F. Hardisty, D.V.M., Chairperson
Experimental Pathology Laboratories, Inc.
D. Dixon, D.V.M., Ph.D.
National Toxicology Program
J. Hellman, D.V.M., Observer
National Toxicology Program
D.E. Malarkey, D.V.M.
National Toxicology Program
R.R. Maronpot, D.V.M.
National Toxicology Program
A. Radovsky, D.V.M., Ph.D.
National Toxicology Program
R.C. Sills, D.V.M., Ph.D.
National Toxicology Program

Biotechnical Services, Inc.
Prepared Technical Report

S.R. Gunnels, M.A., Principal Investigator
L.M. Harper, B.S.
D.C. Serbus, Ph.D.
S.M. Swift, B.S.

CONTENTS

ABSTRACT 5

EXPLANATION OF LEVELS OF EVIDENCE OF CARCINOGENIC ACTIVITY 9

TECHNICAL REPORTS REVIEW SUBCOMMITTEE 10

SUMMARY OF TECHNICAL REPORTS REVIEW SUBCOMMITTEE COMMENTS 11

INTRODUCTION 13

MATERIALS AND METHODS 17

RESULTS 27

DISCUSSION AND CONCLUSIONS 45

REFERENCES 49

APPENDIX A Summary of Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 53

APPENDIX B Summary of Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 99

APPENDIX C Genetic Toxicology 139

APPENDIX D Organ Weights and Organ-Weight-to-Body-Weight Ratios 153

APPENDIX E Hematology Results 155

APPENDIX F Reproductive Toxicity Study Results 157

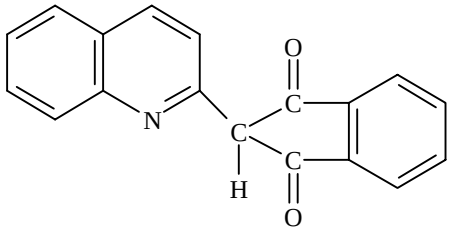
APPENDIX G Chemical Characterization and Dose Formulation Studies 163

APPENDIX H Feed and Compound Consumption in the 2-Year Feed Study of D&C Yellow No. 11 175

APPENDIX I Ingredients, Nutrient Composition, and Contaminant Levels in NIH-07 Rat and Mouse Ration 179

APPENDIX J Sentinel Animal Program 183

ABSTRACT



D&C YELLOW NO. 11

CAS No. 8003-22-3

Chemical Formula: $C_{18}H_{11}NO_2$ Molecular Weight: 273.29

Synonyms: 2-(2-Quinolinyl)-1H-indene-1,3-(2H)-dione; 2-(2-quinolyl)-1,3-indandione
Trade names: Arlosol Yellow S, Chinoline Yellow D (soluble in spirits), Chinoline Yellow ZSS, C.I. 47000, C.I. Solvent Yellow 33, Nitro Fast Yellow SL, Oil Yellow SIS, Petrol Yellow C, Quinoline Yellow A Spirit Soluble, Quinoline Yellow Base, Quinoline Yellow Spirit Soluble, Quinoline Yellow SS, Solvent Yellow 33, Waxoline Yellow T

D&C Yellow No. 11 is used to color topical drug preparations and cosmetics. It is also used in spirit lacquers, polystyrenes, polycarbonates, polyamides, acrylic resins, colored smokes, and hydrocarbon solvents. D&C Yellow No. 11 was nominated to the NTP for toxicity and carcinogenesis studies as part of a larger regulatory effort mandated by Congress and undertaken by the Food and Drug Administration to determine the safety of a number of provisionally listed dyes. D&C Yellow No. 11 is currently regulated for external use. The recommendation to study D&C Yellow No. 11 by dietary exposure was based on the fact that it is a contaminant of D&C Yellow No. 10, a candidate for permanent listing as a chemical for which there is a potential for ingestion.

First-generation (F_0) male and female F344/N rats were given D&C Yellow No. 11 (approximately 99% pure) in feed for up to 19 weeks and then mated, and exposure of second-generation (F_1) males and females began *in utero* and continued for 2 years after weaning at 28 days of age. Genetic toxicology studies were conducted in *Salmonella typhimurium*, cultured Chinese hamster ovary cells, and mouse peripheral blood.

REPRODUCTIVE TOXICITY STUDY

Groups of 60 male and 60 female F_0 rats were given 0, 500, 1,700, or 5,000 ppm D&C Yellow No. 11 in feed for up to 19 weeks, which resulted in average daily doses of 35, 120, or 350 mg D&C Yellow No. 11/kg body weight to males and 35, 120, or 370 mg/kg to females. All F_0 males and females survived until the end of the study. Prior to cohabitation, mean body weight gains of males given 500, 1,700, or 5,000 ppm and of females given 5,000 ppm were significantly lower than those of the controls. The mean body weight gains of exposed females during gestation and lactation were generally similar to those of the controls. Feed consumption by exposed groups of rats was generally similar to that by the control groups prior to cohabitation.

The duration of gestation, the average litter size, the number of live pups on days 4 (precull) and 21, and

the percentage of male pups for each exposure group were similar to those of the controls. The mean body weights of exposed litters were significantly less than those of the control litters on days 14 and 21; this effect was considered to be related to D&C Yellow No. 11 exposure.

2-YEAR STUDY

Groups of 60 male and 60 female F_1 rats were given 0, 500, 1,700, or 5,000 ppm D&C Yellow No. 11 in feed for 105 (males) or 106 (females) weeks after weaning (day 28); 6 to 10 rats per group were evaluated at 12 months. These exposure concentrations resulted in average daily doses of approximately 25, 85, or 250 mg D&C Yellow No. 11/kg body weight to males and 25, 100, or 280 mg/kg to females.

Survival, Body Weights, Feed Consumption, and Clinical Findings

Survival of males given 1,700 or 5,000 ppm was significantly less than that of the controls, and survival of 1,700 ppm females was significantly greater than that of the controls. Mean body weights of 1,700 and 5,000 ppm males and females were generally lower than those of the controls throughout the study. Feed consumption by exposed groups was similar to that by the controls. Chemical-related clinical findings included yellow discoloration of the entire body in all exposed males and females from day 1 and head swelling and edema in 1,700 and 5,000 ppm males. One 1,700 ppm and five 5,000 ppm males were moribund and were killed between weeks 49 and 81; these deaths were attributed to extensive edema.

Hematology

A few minimal hematology changes occurred in male rats at the 12-month interim evaluation. There was evidence of minimal anemia in exposed males; this anemia was characterized by decreased hematocrit values, hemoglobin concentrations, and erythrocyte counts. The minimal anemia was characterized as normocytic, normochromic, and nonresponsive. There were no biologically or statistically significant differences in hematology parameters between control and exposed females.

Pathology Findings

Absolute and relative liver weights of all exposed groups of males and females were significantly greater than those of the controls at 12 months. At 2 years, the incidences of hepatocellular adenoma in 5,000 ppm males and of hepatocellular adenoma or carcinoma (combined) in 5,000 ppm females were significantly greater than those in the controls. At 12 months, the incidences of clear cell foci in 1,700 and 5,000 ppm females were significantly greater than that in the controls. At 2 years, the incidences of mixed cell foci in exposed males and of clear cell foci in exposed males (except 500 ppm) and females were significantly greater than those in the controls. Incidences of cytologic alterations (basophilia and granularity) of hepatocytes, and pigmentation in bile duct epithelium, hepatocytes, and Kupffer cells in exposed males and females were greater than those in the controls at both 12 months and 2 years.

Renal tubule adenomas were observed in two 5,000 ppm males, and one renal tubule carcinoma was observed in a 1,700 ppm male. During an extended evaluation, renal tubule adenomas were observed in two additional 5,000 ppm males, four 1,700 ppm males, and two 500 ppm males. Renal tubule hyperplasia was observed in exposed groups of males but not in controls, and the incidences in 1,700 ppm males from both standard and extended evaluations were significantly greater than those in the controls. Necrosis and regeneration of the renal tubule epithelium were observed in all control and exposed male rats and in most female rats at 12 months and 2 years. The severity of nephropathy in exposed males and females was significantly greater than that in the controls. In exposed males and 1,700 ppm females at 2 years, the incidences of hyperplasia of the transitional epithelium in the kidney, which commonly accompanies advanced nephropathy, were greater than those of the controls, and the severity of this lesion in exposed males and females was greater than that in the controls. The incidences of renal tubule pigmentation in all exposed groups of males and females at 12 months and 2 years were significantly greater than those in the controls.

Squamous cell carcinomas of the tongue were observed in one 500 ppm male at 12 months and one 5,000 ppm female at 2 years, and one squamous cell carcinoma of the oral mucosa was observed in each group of exposed males and in one 5,000 ppm female at 2 years. At 2 years, squamous cell papillomas were observed in the oral cavity (oral mucosa or tongue) of one control, one 500 ppm, two 1,700 ppm, and four 5,000 ppm

males; this lesion was also observed in one control and one 500 ppm female.

GENETIC TOXICOLOGY

Results of mutagenicity tests with D&C Yellow No. 11 in *Salmonella typhimurium* were equivocal in one study, based on responses observed in strain TA100 with induced rat liver S9, and weakly positive in a second study, based on responses observed in strains TA98 and TA100 with induced rat or hamster liver S9. D&C Yellow No. 11 induced sister chromatid exchanges and chromosomal aberrations in cultured Chinese hamster ovary cells, with and without S9. No increase in the frequency of micronucleated normochromatic erythrocytes was observed in peripheral blood samples from male and female $B6C3F_1$ mice administered D&C Yellow No. 11 in feed for 13 weeks.

CONCLUSIONS

Under the conditions of this perinatal exposure followed by a 2-year dosed feed study, there was *some evidence of carcinogenic activity** of D&C Yellow No. 11 in male F344/N rats based on increased incidences of hepatocellular adenoma, renal tubule neoplasms, and squamous cell neoplasms of the oral cavity. There was *some evidence of carcinogenic activity* in female F344/N rats based on increased incidences of hepatocellular neoplasms. Incidences of uncommon squamous cell carcinoma of the oral cavity in females may have been related to chemical treatment.

Exposure of rats to D&C Yellow No. 11 in feed for 2 years resulted in increased incidences of nonneoplastic liver lesions including clear cell foci, increased basophilia and granularity in the cytoplasm of hepatocytes, and bile duct, hepatocyte, and Kupffer cell pigmentation in males and females and mixed cell foci in males. In the kidney, there were increased incidences of renal tubule pigmentation and transitional epithelial hyperplasia in males and females and renal tubule hyperplasia in males. The severity of nephropathy was increased in exposed males and females.

---

* Explanation of Levels of Evidence of Carcinogenic Activity is on page 9. A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 11.

Summary of the 2-Year Carcinogenesis and Genetic Toxicology Studies of D&C Yellow No. 11

	Male F344/N Rats	Female F344/N Rats
Doses	0, 500, 1,700, or 5,000 ppm	0, 500, 1,700, or 5,000 ppm
Body weights	1,700 and 5,000 ppm groups lower than control group	1,700 and 5,000 ppm groups lower than control group
2-Year survival rates	19/50, 20/51, 8/51, 2/54	22/50, 26/51, 37/50, 23/51
Nonneoplastic effects	Liver: clear cell focus (9/50, 15/51, 15/51, 18/54); mixed cell focus (1/50, 10/51, 9/51, 10/54); bile duct pigmentation (0/50, 38/51, 51/51, 54/54); hepatocyte cytologic alterations (0/50, 20/51, 44/51, 42/54); hepatocyte pigmentation (0/50, 22/51, 45/51, 51/54); Kupffer cell pigmentation (7/50, 15/51, 23/51, 26/54) Kidney: renal tubule hyperplasia (standard evaluation – 0/50, 0/51, 4/51, 3/54; extended evaluation – 0/50, 2/51, 9/51, 2/54; standard and extended evaluations combined – 0/50, 2/51, 13/51, 4/54); renal tubule pigmentation (18/50, 43/51, 47/51, 54/54); transitional epithelial hyperplasia (11/50, 23/51, 29/51, 34/54); severity of nephropathy (2.3, 2.8, 3.2, 3.0)	Liver: clear cell focus (10/50, 18/51, 29/50, 30/51); bile duct pigmentation (0/50, 46/51, 49/50, 50/51); hepatocyte cytologic alterations (0/50, 11/51, 31/50, 40/51); hepatocyte pigmentation (0/50, 34/51, 44/50, 50/51); Kupffer cell pigmentation (9/50, 11/51, 16/50, 32/51) Kidney: renal tubule pigmentation (10/50, 48/51, 50/50, 51/51); transitional epithelial hyperplasia (2/50, 6/51, 10/50, 3/51); severity of nephropathy (1.4, 1.7, 1.8, 2.1)
Neoplastic effects	Liver: hepatocellular adenoma (1/50, 2/51, 1/51, 7/54) Kidney: renal tubule adenoma (standard evaluation – 0/50, 0/51, 0/51, 2/54; extended evaluation – 0/50, 2/51, 4/51, 2/54; standard and extended evaluations combined – 0/50, 2/51, 4/51, 4/54); renal tubule adenoma or carcinoma (standard and extended evaluations combined – 0/50, 2/51, 5/51, 4/54) Oral cavity: squamous cell papilloma (1/50, 1/51, 2/51, 4/54); squamous cell carcinoma (0/50, 1/51, 1/51, 1/54); squamous cell papilloma or squamous cell carcinoma (1/50, 2/51, 3/51, 5/54)	Liver: hepatocellular adenoma or carcinoma (0/50, 2/51, 5/50, 5/51)
Uncertain finding	None	Oral cavity: squamous cell carcinoma (0/50, 0/51, 0/50, 2/51); squamous cell papilloma or squamous cell carcinoma (1/50, 1/51, 0/50, 2/51)
Level of evidence of carcinogenic activity	Some evidence	Some evidence

Genetic toxicology

Salmonella typhimurium gene mutations:	Equivocal in strain TA100 with S9 at SRI International, and weakly positive in strains TA98 and TA100 with S9 at Microbiological Associates, Inc.
Sister chromatid exchanges	
Cultured Chinese hamster ovary cells *in vitro*:	Positive with and without S9
Chromosomal aberrations	
Cultured Chinese hamster ovary cells *in vitro*:	Positive with and without S9
Micronucleated erythrocytes	
Mouse peripheral blood *in vivo*:	Negative

EXPLANATION OF LEVELS OF EVIDENCE OF CARCINOGENIC ACTIVITY

The National Toxicology Program describes the results of individual experiments on a chemical agent and notes the strength of the evidence for conclusions regarding each study. Negative results, in which the study animals do not have a greater incidence of neoplasia than control animals, do not necessarily mean that a chemical is not a carcinogen, inasmuch as the experiments are conducted under a limited set of conditions. Positive results demonstrate that a chemical is carcinogenic for laboratory animals under the conditions of the study and indicate that exposure to the chemical has the potential for hazard to humans. Other organizations, such as the International Agency for Research on Cancer, assign a strength of evidence for conclusions based on an examination of all available evidence, including animal studies such as those conducted by the NTP, epidemiologic studies, and estimates of exposure. Thus, the actual determination of risk to humans from chemicals found to be carcinogenic in laboratory animals requires a wider analysis that extends beyond the purview of these studies.

Five categories of evidence of carcinogenic activity are used in the Technical Report series to summarize the strength of the evidence observed in each experiment: two categories for positive results (**clear evidence** and **some evidence**); one category for uncertain findings (**equivocal evidence**); one category for no observable effects (**no evidence**); and one category for experiments that cannot be evaluated because of major flaws (**inadequate study**). These categories of interpretative conclusions were first adopted in June 1983 and then revised in March 1986 for use in the Technical Report series to incorporate more specifically the concept of actual weight of evidence of carcinogenic activity. For each separate experiment (male rats, female rats, male mice, female mice), one of the following five categories is selected to describe the findings. These categories refer to the strength of the experimental evidence and not to potency or mechanism.

- **Clear evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a dose-related (i) increase of malignant neoplasms, (ii) increase of a combination of malignant and benign neoplasms, or (iii) marked increase of benign neoplasms if there is an indication from this or other studies of the ability of such tumors to progress to malignancy.
- **Some evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a chemical-related increased incidence of neoplasms (malignant, benign, or combined) in which the strength of the response is less than that required for clear evidence.
- **Equivocal evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a marginal increase of neoplasms that may be chemical related.
- **No evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing no chemical-related increases in malignant or benign neoplasms.
- **Inadequate study** of carcinogenic activity is demonstrated by studies that, because of major qualitative or quantitative limitations, cannot be interpreted as valid for showing either the presence or absence of carcinogenic activity.

When a conclusion statement for a particular experiment is selected, consideration must be given to key factors that would extend the actual boundary of an individual category of evidence. Such consideration should allow for incorporation of scientific experience and current understanding of long-term carcinogenesis studies in laboratory animals, especially for those evaluations that may be on the borderline between two adjacent levels. These considerations should include:

- adequacy of the experimental design and conduct;
- occurrence of common versus uncommon neoplasia;
- progression (or lack thereof) from benign to malignant neoplasia as well as from preneoplastic to neoplastic lesions;
- some benign neoplasms have the capacity to regress but others (of the same morphologic type) progress. At present, it is impossible to identify the difference. Therefore, where progression is known to be a possibility, the most prudent course is to assume that benign neoplasms of those types have the potential to become malignant;
- combining benign and malignant tumor incidence known or thought to represent stages of progression in the same organ or tissue;
- latency in tumor induction;
- multiplicity in site-specific neoplasia;
- metastases;
- supporting information from proliferative lesions (hyperplasia) in the same site of neoplasia or in other experiments (same lesion in another sex or species);
- presence or absence of dose relationships;
- statistical significance of the observed tumor increase;
- concurrent control tumor incidence as well as the historical control rate and variability for a specific neoplasm;
- survival-adjusted analyses and false positive or false negative concerns;
- structure-activity correlations; and
- in some cases, genetic toxicology.

NATIONAL TOXICOLOGY PROGRAM BOARD OF SCIENTIFIC COUNSELORS
TECHNICAL REPORTS REVIEW SUBCOMMITTEE

The members of the Technical Reports Review Subcommittee who evaluated the draft NTP Technical Report on D&C Yellow No. 11 on 5 December 1995, are listed below. Subcommittee members serve as independent scientists, not as representatives of any institution, company, or governmental agency. In this capacity, subcommittee members have five major responsibilities in reviewing NTP studies:

- to ascertain that all relevant literature data have been adequately cited and interpreted,
- to determine if the design and conditions of the NTP studies were appropriate,
- to ensure that the Technical Report presents the experimental results and conclusions fully and clearly,
- to judge the significance of the experimental results by scientific criteria, and
- to assess the evaluation of the evidence of carcinogenic activity and other observed toxic responses.

Arnold L. Brown, M.D., Chairperson
University of Wisconsin Medical School
Madison, WI

Gary P. Carlson, Ph.D., Principal Reviewer
School of Health Sciences
Purdue University
West Lafayette, IN

Thomas L. Goldsworthy, Ph.D.
Department of Experimental Pathology and Toxicology
Chemical Industry Institute of Toxicology
Research Triangle Park, NC

Robert LeBoeuf, Ph.D.
Corporate Professional and Regulatory Services
Human Safety Department
The Procter & Gamble Company
Cincinnati, OH

Janardan K. Reddy, M.D., Principal Reviewer*
Department of Pathology
Northwestern University Medical School
Chicago, IL

Irma Russo, M.D., Principal Reviewer
Fox Chase Cancer Center
Philadelphia, PA

Louise Ryan, Ph.D.
Division of Biostatistics
Dana-Farber Cancer Institute
Boston, MA

Robert E. Taylor, M.D., Ph.D.
Department of Pharmacology
Howard University College of Medicine
Washington, DC

Frederick L. Tyson, Ph.D.
St. Mary's Hospital and Medical Center
Cancer Research Institute
Grand Junction, CO

Jerrold M. Ward, D.V.M., Ph.D.*
National Cancer Institute
Frederick, MD

---

* Did not attend

SUMMARY OF TECHNICAL REPORTS REVIEW SUBCOMMITTEE COMMENTS

On 5 December 1995, the draft Technical Report on the toxicology and carcinogenesis studies of D&C Yellow No. 11 received public review by the National Toxicology Program's Board of Scientific Counselors' Technical Reports Review Subcommittee. The review meeting was held at the National Institute of Environmental Health Sciences, Research Triangle Park, NC.

Dr. W.C. Eastin, NIEHS, introduced the toxicology and carcinogenesis studies of D&C Yellow No. 11 by discussing the uses of the chemical and rationale for study, describing the experimental design, reporting on survival and body weight effects, and commenting on chemical-related neoplasms and nonneoplastic lesions in male and female rats. Dr. Eastin reported that the study of this color additive was part of a larger effort mandated by Congress and undertaken by the FDA to determine the safety of provisionally listed dyes. The study design was not a standard NTP protocol. In discussions with the nominator, FDA, NTP decided to tailor the protocol to provide perinatal exposure followed by dietary exposure for 2 years in order to generate data similar to those used by the FDA to regulate other color additives. The proposed conclusions were *some evidence of carcinogenic activity* in male and female F344/N rats.

Dr. Reddy, a principal reviewer, was unable to attend the meeting but had submitted his review, which Dr. L.G. Hart, NIEHS, read into the record. Dr. Reddy agreed with the proposed conclusions. He said the abstract should give the reasons for using only rats in this study.

Dr. Russo, the second principal reviewer, agreed with the proposed conclusions. She also thought there needed to be clarification of why concurrent studies were not done in mice. Dr. Eastin said that in prechronic studies effects in mice were about the same as in rats, although in all endpoints measured, rats were the more sensitive species. To have done both species with the larger perinatal protocol would have diverted resources from studying another chemical. Conducting the study in the more sensitive species would meet the FDA's needs.

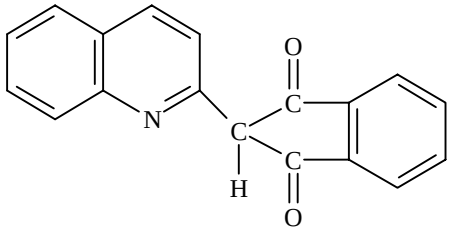
Dr. Carlson, the third principal reviewer, agreed with the proposed conclusions. He commented that he disagreed with the perinatal protocol from this and other such studies and claims made about effects *in utero*, particularly when there are no groups exposed only post-weaning for comparison. Dr. Eastin agreed that these groups would have been useful as well as animals exposed only *in utero*. Further, Dr. Carlson said the discussion mentions positive findings, therefore, negative findings should be cited. Dr. Eastin reported that there are only three other NTP studies with prenatal or perinatal exposures. Dr. Carlson said he was intrigued by the description of head swelling and edema and asked for more information on etiology. Dr. A. Radovsky, NIEHS, said the possibilities of hypoproteinemia, secondary to kidney or liver disease or intestinal malabsorption, and vascular or heart lesions were investigated. All of these conditions were present in some but not in all animals with edema, and the severity of kidney or liver neoplasms was not any greater in these animals than in cohorts without edema. Thus, from an anatomic histopathologic perspective, there was no explanation.

Dr. LeBoeuf noted a body weight reduction in 5,000 ppm males of about 15% and wondered if this was typical or acceptable for NTP studies, rather than the 10%, which he thought was associated with reaching a maximum tolerated dose. Dr. J.R. Bucher, NIEHS, responded that it depends on the study outcome. If there is a neoplasm response in a study that has a 15% decrease, that would be acceptable, whereas in a negative study, such a large decrease might help prevent development of a neoplastic response.

Dr. Bucher reported that in 5,000 ppm female rats, a second unusual oral cavity carcinoma was observed. Thus, NTP proposed adding a sentence to the end of the first paragraph of the conclusions, but the primary level of evidence in female rats would remain *some evidence of carcinogenic activity*.

Dr. Russo moved that the Technical Report on D&C Yellow No. 11 be accepted with the revisions discussed and the conclusions as written for male and female rats. Dr. Carlson seconded the motion, which was accepted with six yes votes and one abstention (Dr. LeBoeuf).

INTRODUCTION



D&C YELLOW NO. 11

CAS No. 8003-22-3

Chemical Formula: $C_{18}H_{11}NO_2$ Molecular Weight: 273.29

Synonyms: 2-(2-Quinolinyl)-1H-indene-1,3-(2H)-dione; 2-(2-quinolyl)-1,3-indandione
Trade names: Arlosol Yellow S, Chinoline Yellow D (soluble in spirits), Chinoline Yellow ZSS, C.I. 47000, C.I. Solvent Yellow 33, Nitro Fast Yellow SL, Oil Yellow SIS, Petrol Yellow C, Quinoline Yellow A Spirit Soluble, Quinoline Yellow Base, Quinoline Yellow Spirit Soluble, Quinoline Yellow SS, Solvent Yellow 33, Waxoline Yellow T

CHEMICAL AND PHYSICAL PROPERTIES

D&C Yellow No. 11 is a bright greenish yellow solid or a canary yellow powder with a melting point range of 240.9° to 242.1° C. It is soluble in acetone, benzene, chloroform, toluene, and xylene; slightly soluble in methanol, ethanol, ethyl acetate, linseed oil, mineral oil, oleic acid, paraffin wax, stearic acid, and turpentine; and insoluble in water (*Colour Index*, 1982; *Merck Index*, 1989). D&C Yellow No. 11 is the name given to 2-(2-quinolyl)-1,3-indandione when it meets United States Certification Regulations (21 CFR, §74.1711). These regulations state that the certified dye must conform to the following specifications and be free from impurities other than those named: volatile matter ≤ 1%, matter insoluble in ethyl alcohol ≤ 0.4%, phthalic acid ≤ 0.3%, quinaldine ≤ 0.2%, subsidiary colors ≤ 5%, lead ≤ 20 ppm, arsenic ≤ 3 ppm, and mercury ≤ 1 ppm. D&C Yellow No. 11 does not contain the methylated congener 6-methyl-2-(2-quinolyl)-1,3-indandione. However, the noncertified dye, usually referred to as Solvent Yellow 33 (CTFA, 1982), is composed of two parts nonmethylated and one part methylated forms of the dye (*Colour Index*, 1982). In some toxicity studies, the D&C Yellow No. 11 that was used contained both 2-(2-quinolyl)-1,3-indandione and 6-methyl-2-(2-quinolyl)-1,3-indandione (Björkner and Niklasson, 1983; Weaver, 1983; Sato *et al.*, 1984).

PRODUCTION, USE, AND HUMAN EXPOSURE

D&C Yellow No. 11 is generally used in solvent form to color topical drug preparations and cosmetics (CTFA, 1982; El Dareer *et al.*, 1988). In the United States, D&C Yellow No. 11 is approved only for external applications (21 CFR, §74.1711; Marmion, 1991). It is also used in spirit lacquers, polystyrenes, polycarbonates, polyamides, acrylic resins, colored smokes, and occasionally hydrocarbon solvents (*Merck Index*, 1989). Between 1985 and 1995, 24,580 pounds of D&C Yellow No. 11 were certified, and 131 cosmetic formulations containing the dye were reported (FDA, personal communication). The National Occupational Exposure Survey estimated that 14,313 workers (4,310 females) were potentially exposed to D&C Yellow No. 11 in five different industries from 1981 to 1983 (NIOSH, 1990).

ABSORPTION, DISTRIBUTION, METABOLISM, AND EXCRETION

Experimental Animals

Studies sponsored by the NTP demonstrated that radiolabeled D&C Yellow No. 11 was rapidly absorbed, distributed, and excreted by male F344/N rats (El Dareer *et al.*, 1988). D&C Yellow No. 11 was not concentrated to any great extent in any tissue (range 0.006% to 0.88% per tissue 72 hours following intravenous, oral, or repeated oral administration) and was rapidly excreted in feces following both oral and intravenous administration. On a µg Eq/g tissue basis, the concentration of D&C Yellow No. 11-derived radioactivity 72 hours after administration was approximately an order of magnitude greater in the liver and kidney than in the other tissues. Excretion in the feces accounted for approximately 80% of an intravenous dose within 24 hours of administration and 85% within 72 hours. Most of the remainder of the dose was detected in the urine within 24 hours with only trace amounts, approximately 2%, remaining in the tissues 72 hours after dosing. Results of bile cannulation studies indicated that excretion in feces is the result of rapid metabolism and excretion in bile (El Dareer *et al.*, 1988). Greater than 50% of an intravenous dose was excreted in bile within 4 hours of administration.

Humans

No information on the absorption, distribution, metabolism, or excretion of D&C Yellow No. 11 in humans was found in the literature.

TOXICITY

Experimental Animals

No deaths occurred in albino rats (strain not given) given 2,500 to 50,000 ppm D&C Yellow No. 11 in feed for 13 weeks; however, the liver was enlarged at all concentrations studied (Hansen *et al.*, 1960).

In a study by Sun *et al.* (1987), F344/N rats were exposed to Solvent Yellow 33 aerosol by inhalation 6 hours per day, 5 days per week, for 14 days or 13 weeks. After 14 days of exposure (10, 51, or 230 mg/m^3), rats exposed to 230 mg/m^3 had body weights 8% lower than those of the controls. After 13 weeks of exposure (1, 10.8, or 100 mg/m^3), rats exposed to 100 mg/m^3 had body weights 5% lower than those of the controls and an accumulation of vacuolated alveolar macrophages in the lung. However, tissue analysis by high-performance liquid chromatography showed very little Solvent Yellow 33 in the lung after exposure, indicating rapid clearance.

In toxicity studies conducted by the NTP, D&C Yellow No. 11 (approximately 99% pure) was administered in feed to male and female F344/N rats and B6C3F$_1$ mice at concentrations up to 50,000 ppm for 14 days (five animals per group) or 13 weeks (10 animals per group) (NTP, 1991a). Although the estimated intake of D&C Yellow No. 11 by mice was more than twice that by rats, the results of the 14-day and 13-week studies were similar for both species. No deaths occurred in rats or mice in the 14-day or 13-week studies, but body weight gains were slightly reduced in male and female rats given 17,000 or 50,000 ppm. Liver weights of exposed rats and mice were greater than those of controls. There was minimal to mild degeneration of the periportal portion of the liver lobules of rats given 1,700 ppm or greater and of mice given 5,000 ppm or greater. A dose-related yellow-brown pigment was observed in the hepatocytes, Kupffer cells, and biliary epithelium in male and female rats and mice and in the renal tubule epithelium in male and female rats. Hepatocellular degeneration progressed slightly in severity with increased time of exposure (14 days versus 13 weeks) in rats but not in mice. Cytoplasmic alteration, an increase in the size and number of hyaline droplets, in the renal tubule epithelium of the cortex and outer medulla was present in all exposed groups of male rats. The conclusions from these studies were that D&C Yellow No. 11 caused increased liver weights in male and female rats and mice and increases in the size and number of hyaline droplets in male rats at all exposure concentrations.

Because of the cytoplasmic alteration (protein droplet accumulation) observed in male rats given D&C Yellow No. 11 in the 13-week NTP study (NTP, 1991a), additional studies were conducted to determine the potential for regression of these chemical-related lesions (Eastin *et al.*, 1996). Groups of six male rats given feed containing 5,000 ppm D&C Yellow No. 11 or untreated feed for 70 days, then maintained on undosed feed, were examined the last day of exposure (day 1) and on days 3, 14, and 28 of the recovery

period. On day 1, cytoplasmic alteration and pigment in the renal tubules and hepatocellular degeneration and pigmentation were similar to the lesions observed at the same exposure concentration in the 13-week study. After a recovery period of 3 days, the severities of cytoplasmic alteration and pigmentation in the renal tubule epithelium were reduced in all rats. At this time, there was no longer morphologic evidence of the hepatocellular degeneration, and although the pigmentation was slightly less prominent, it was still present in the biliary epithelium and cytoplasm of hepatocytes and Kupffer cells in the periportal areas. After recovery periods of 14 or 28 days, pigment was still present in the renal tubule epithelium and liver biliary epithelium of all exposed rats. Ultrastructural features included an electron-dense, homogenous pigment in the cytoplasm of canaliculi, bile duct epithelium, and the lumen of bile ducts. Protein droplet accumulation resembled α2μ-globulin by light microscopy; however, there was no evidence of an increase in the amount of α2μ-globulin (as percent of total protein) measured by an ELISA method (Charbonneau *et al.*, 1987; Yuan *et al.*, 1992) in the kidney of rats with cytoplasmic alteration. When measured on day 1, the amount of α2μ-globulin in the kidney of control and exposed rats was 10.0% and 8.1%, respectively. On days 3, 14, and 28, these values were similar in exposed and control groups.

When partially hepatectomized Charles River male rats were given 15,000 ppm D&C Yellow No. 11 in feed for 10 days after surgery, liver regeneration was stimulated significantly compared with that in partially hepatectomized controls (Gershbein, 1982).

D&C Yellow No. 11 was shown to sensitize the skin of adult Hartley guinea pigs. Females induced with 40% D&C Yellow No. 11 in ethanol with a 24-hour occluded patch once a week for 3 consecutive weeks responded to a challenge concentration of 10% administered after a 2-week rest period (Lamson *et al.*, 1982). Hartley guinea pigs were also induced by injection of emulsified Freund's complete adjuvant into the nuchal region, followed by application of one of four test samples of D&C Yellow No. 11 to abraded skin for 2 days and topical application on days 8 and 9 (Sato *et al.*, 1984). Challenge was carried out by topical application on day 21 to flank skin. In these studies, the threshold concentration for induction and challenge was 10 ppm. In another study, D&C Yellow No. 11 in Freund's adjuvant injected into the footpad produced dose-response hypersensitivity in female Hartley guinea pigs 2 weeks after exposure to intradermal challenges of the dye (Palazzolo and DiPasquale, 1983). Histopathologic examination of reaction sites indicated a cellular inflammatory response in guinea pigs consistent with delayed-type hypersensitivity.

Humans

D&C Yellow No. 11 has been shown to have a high allergenic potential in humans (Kita *et al.*, 1984). Patients sensitized to D&C Yellow No. 11 in maximization tests exhibited an allergic contact dermatitis from the use of soaps (Jordan, 1981; Weaver, 1983) and facial cosmetics (Björkner and Magnusson, 1981; Calnan, 1981; Björkner and Niklasson, 1983; Rapaport, 1984) containing this dye. Positive reactions were seen in beauticians with hand dermatitis given Quinoline Yellow SS (0.5% in petrolatum) (Matsunaga *et al.*, 1988).

REPRODUCTIVE AND DEVELOPMENTAL TOXICITY

Experimental Animals

In a perinatal exposure study, body weight gains of rat dams given diets containing 5,000, 17,000, or 50,000 ppm D&C Yellow No. 11 for 4 weeks before mating to unexposed males were similar to that of the controls at the time of mating but were lower than those of the controls at parturition and at weaning (NTP, 1991a). However, fertility, length of gestation, litter size, and pup birth body weights were unaffected by exposure. At weaning, body weights of pups from all exposed dams were lower than those from control dams. After potential exposure to D&C Yellow No. 11 for 4 weeks through the milk and subsequently in feed at the same concentrations given their dams, body weights of male and female pups given 5,000 ppm were similar to those of the controls, but those of pups given 17,000 or 50,000 ppm were lower than those of the controls. Microscopic evaluation showed lesions in pups from all exposure groups; these lesions were similar to those in the liver and kidney of rats in the 14-day and 13-week studies (NTP, 1991a). In the liver, degeneration of hepatocytes was present in all exposed groups and was characterized by minimal cytoplasmic vacuolization.

All exposed rats had a minimal accumulation of a granular-to-globular yellow-brown pigment in the cytoplasms of cells in the liver and kidney. In the kidney of exposed males, there was cytoplasmic alteration (hyaline droplets) similar to that observed in the males in the 14-day and 13-week studies (NTP, 1991a; Eastin *et al.*, 1996).

Humans

No information on the reproductive or developmental toxicity of D&C Yellow No. 11 in humans was found in the literature.

CARCINOGENICITY

No information on the carcinogenic potential of D&C Yellow No. 11 in experimental animals or in humans was found in the literature.

GENETIC TOXICITY

D&C Yellow No. 11 has been shown to be mutagenic *in vitro*. It induced mutations in *Salmonella typhimurium* strains TA98 and TA100 when exposure occurred in the presence of S9 metabolic activation enzymes (Table C1; Zeiger *et al.*, 1988). In a second study, mutations were induced in *S. typhimurium* strains TA102 and TA104 with and without S9 (Moore *et al.*, 1988). D&C Yellow No. 11 was also mutagenic and clastogenic to L5178Y/TK mouse lymphoma cells with and without S9 (Meyer *et al.*, 1986; Moore *et al.*, 1988). Sister chromatid exchange levels were also elevated in mouse lymphoma cells treated with D&C Yellow No. 11 in the presence of S9 (Moore *et al.*, 1988). In contrast to the demonstrated *in vitro* mutagenicity of D&C Yellow No. 11 in a number of assays, no increase in the frequency of sister chromatid exchanges was observed *in vivo* in bone marrow cells of male mice administered a single intraperitoneal injection of 10, 20, or 40 mg D&C Yellow No. 11/kg body weight (Moore *et al.*, 1988).

STUDY RATIONALE

D&C Yellow No. 11 was nominated to the NTP for toxicity and carcinogenesis studies as part of a larger regulatory effort mandated by Congress and undertaken by the FDA to determine the safety of a number of provisionally listed dyes. Currently, D&C Yellow No. 11 is regulated by the FDA for external use only (21 CFR, §74.1711). The decision to obtain toxicity and carcinogenesis data for this color additive by dietary exposure studies was based on the fact that it is a contaminant of D&C Yellow No. 10, a color additive approved for internal use and a candidate for permanent listing. The toxic effects of oral exposure to D&C Yellow No. 11 were unknown in mice and had not been determined in rats in 2-year studies. Thus, 14-day and 13-week toxicity studies were conducted in F344/N rats and B6C3F$_1$ mice (NTP, 1991a) in order to compare the results to the NTP historical database for these strains. These studies were reported at the time of their completion to the National Toxicology Program's Board of Scientific Counselors' Technical Reports Review Subcommittee. However, the FDA only requires carcinogenesis study data from one rodent species, and different rat strains have been used the most, primarily the Sprague-Dawley rat. NTP also selected the rat based on the fact that the toxic effects in rats and mice after 13 weeks of dietary exposure to D&C Yellow No. 11 were basically the same, and rats were slightly more sensitive than mice. NTP also used the F344/N rat to be able to compare the results of the carcinogenicity study with their large historical database on this strain.

MATERIALS AND METHODS

PROCUREMENT AND CHARACTERIZATION OF D&C YELLOW NO. 11

D&C Yellow No. 11 was obtained from H. Kohnstamm and Company, Inc. (New York), in one lot (ZB2016) and certified by the Food and Drug Administration, Division of Color Technology. Identity, purity, and stability analyses were conducted by the analytical chemistry laboratory, Midwest Research Institute (Kansas City, MO) (Appendix G). Reports on analyses performed in support of the D&C Yellow No. 11 studies are on file at the National Institute of Environmental Health Sciences (NIEHS).

The chemical, a yellow powder, was identified as D&C Yellow No. 11 by infrared, ultraviolet/visible, nuclear magnetic resonance, and direct inlet mass spectrometry. Purity was determined by elemental analyses, Karl Fischer water analysis, thin-layer chromatography, and high-performance liquid chromatography. Elemental analyses for carbon, hydrogen, and nitrogen were in agreement with the theoretical values for D&C Yellow No. 11. Karl Fischer water analysis indicated less than 0.02% water. Thin-layer chromatography indicated one major spot by one system and one major spot and one trace impurity by a second system. High-performance liquid chromatography revealed a major peak and two impurities with areas greater than 0.1% of the major peak area. The overall purity was determined to be approximately 99%.

Stability studies of the bulk chemical were performed using high-performance liquid chromatography. These studies indicated that D&C Yellow No. 11 was stable as a bulk chemical for 2 weeks when stored protected from light at temperatures up to 60 °C. To ensure stability, the bulk chemical was stored at room temperature in its original packaging protected from light. Stability was monitored during the reproductive toxicity and 2-year studies using high-performance liquid chromatography. No degradation of the bulk chemical was detected.

PREPARATION AND ANALYSIS OF DOSE FORMULATIONS

The dose formulations were prepared every 2 weeks by mixing D&C Yellow No. 11 with feed (Table G1). Homogeneity and stability studies of the 500 ppm dose formulation were performed by the analytical chemistry laboratory using high-performance liquid chromatography. Homogeneity was confirmed and the stability of the dose formulations was confirmed for at least 3 weeks when stored protected from light at room temperature and for 7 days when stored open to air and light.

Periodic analyses of the dose formulations of D&C Yellow No. 11 were conducted at the study laboratory using visible spectrometry. During the reproductive toxicity and 2-year studies, the formulations were analyzed approximately every 8 weeks (Table G2). All of the dose formulations used in the studies were within 10% of the target concentration. Due to an unacceptable ratio of duplicate analyses, one dose formulation was remixed. Results of a referee analysis performed by the analytical chemistry laboratory agreed with the results obtained by the study laboratory (Table G3).

REPRODUCTIVE TOXICITY STUDY

The reproductive toxicity study was conducted to evaluate the cumulative toxic effects of parental and *in utero* exposure to D&C Yellow No. 11; pups from this study continued to receive dosed feed at the same concentrations as their dams for the 2-year study.

Thirty-two-day-old male and female F344/N first-generation (F_0) rats were obtained from Taconic Farms (Germantown, NY) and quarantined for 10 days before receiving test diets. Rats were 112 days old on the first day of cohabitation. Before initiation of the study, five male and five female rats were randomly selected for parasite evaluation and gross observation for evidence of disease. At the end of the study, serologic analyses were performed on five male and five female

control rats using the protocols of the NTP Sentinel Animal Program (Appendix J).

Groups of 60 male and 60 female rats were fed diets containing 0, 500, 1,700, or 5,000 ppm D&C Yellow No. 11 beginning 10 weeks prior to cohabitation, during cohabitation, and through gestation and lactation (females). Feed and water were available *ad libitum*. Rats were housed five per cage except during cohabitation (one male and one female per cage) and lactation (one dam and litter per cage). Clinical findings, feed consumption, and body weights were recorded on day 1 (feed consumption day 2), once per week before cohabitation, on days 0, 6, 15, and 21 of gestation (females), and on days 1, 4, 14, and 21 of lactation (females and pups). Details of the study design and animal maintenance are summarized in Table 1; the following timeline describes the exposure periods.

First-generation (F_0) rats started on D&C Yellow No. 11 in feed

⇓

70-day precohabitation period

⇓

7-day cohabitation period

⇓

21-day gestation period

⇓

28-day lactation period

⇓

Second-generation (F_1) rats continued on same concentrations of D&C Yellow No. 11 in feed as dams for 2 years

⇓

Terminal sacrifice

During cohabitation, vaginal smears were taken daily from females to determine the presence of sperm. Rats showing no signs of littering by day 25 were killed, and uteri were examined for evidence of unsuccessful pregnancy. If there was no gross evidence of pregnancy, uteri were stained with ammonium sulfide or sodium sulfide and examined for implantation sites. After parturition, clinical signs and number and sex of live pups were recorded. On day 4 postpartum, litters were randomly culled to a maximum of eight pups (four male and four female) per litter; on day 21, 60 male and 60 female pups were randomly selected from the litters of each exposure group, and these pups were weaned on day 28 and continued on the same test diet as their dams. Clinical findings and pup weights were recorded on days 1, 4, 14, and 21.

2-YEAR STUDY

Study Design

Groups of 60 male and 60 female second-generation (F_1) rats were fed diets containing 0, 500, 1,700, or 5,000 ppm D&C Yellow No. 11 for 105 to 106 weeks. Up to 10 male and 10 female rats from each group were evaluated at 12 months for hematology, organ weights, and histopathology.

Source and Specification of Animals

Male and female F_1 rats were selected from litters produced by breeding male and female F344/N rats in-house after exposure to 0, 500, 1,700, or 5,000 ppm D&C Yellow No. 11 in feed for 70 days. Rats were monitored for parasites throughout the study. Rats were 28 days old when weaned at the beginning of the study. The health of the animals was monitored during the studies according to the protocols of the NTP Sentinel Animal Program (Appendix J).

Animal Maintenance

Rats were housed five per cage. Feed and water were available *ad libitum*. Feed consumption was measured during week 2 and at monthly intervals thereafter by cage (Appendix H). Cages and racks were rotated every 2 weeks. Further details of animal maintenance are given in Table 1. Information on feed composition and contaminants is provided in Appendix I.

Clinical Examinations and Pathology

All animals were observed twice daily. Clinical findings and body weights were recorded at least once a week for the first 13 weeks and every 4 weeks thereafter.

A complete necropsy and microscopic examination were performed on all rats. At the 12-month interim evaluation, the liver and right kidney were weighed. At necropsy, all organs and tissues were examined for grossly visible lesions, and all major tissues were fixed and preserved in 10% neutral buffered formalin, processed and trimmed, embedded in paraffin, sectioned to a thickness of approximately 5 µm, and stained with hematoxylin and eosin for microscopic examination. For all paired organs (i.e., adrenal gland, kidney, ovary), samples from each organ were examined. For extended evaluation of renal proliferative lesions, kidneys were step sectioned at 1 mm intervals, and four additional sections were obtained from each kidney. Tissues examined microscopically are listed in Table 1.

Hematology studies were performed on up to 10 male and 10 female rats per group at the 12-month interim evaluation. Rats were anesthetized with a CO_2/O_2 mixture, and blood was drawn from the retroorbital sinus. Blood for hematology determinations was placed in tubes containing potassium ethylenediaminetetraacetic acid as an anticoagulant. The hematology variables evaluated are listed in Table 1. Erythrocyte and leukocyte counts, hemoglobin concentration, hematocrit, mean cell volume, mean cell hemoglobin, mean cell hemoglobin concentration, and platelet counts were performed on a Technicon H-1 hematology analyzer (Tarrytown, NY). Differential leukocyte counts, morphologic evaluation of blood cells, and nucleated erythrocyte counts were determined by light microscopy using smears prepared from blood stained by incubating equal volumes of whole blood and new methylene blue for at least 20 minutes.

Microscopic evaluations were completed by the study laboratory pathologist, and the pathology data were entered into the Toxicology Data Management System. The microscopic slides, paraffin blocks, and residual wet tissues were sent to the NTP Archives for inventory, slide/block match, and wet tissue audit. The slides, individual animal data records, and pathology tables were evaluated by an independent quality assessment laboratory. The individual animal records and tables were compared for accuracy, the slide and tissue counts were verified, and the histotechnique was evaluated. For the 2-year study, a quality assessment pathologist reviewed the forestomach, kidney, liver, lung, lymph nodes, salivary glands, and spleen of males and females; the mammary gland, oral mucosa, pancreas, parathyroid gland, small intestine, and tongue of males; and the clitoral gland of females.

The quality assessment report and the reviewed slides were submitted to the NTP Pathology Working Group (PWG) chairperson, who reviewed the selected tissues and addressed any inconsistencies in the diagnoses made by the laboratory and quality assessment pathologists. Representative histopathology slides containing examples of lesions related to chemical administration, examples of disagreements in diagnoses between the laboratory and quality assessment

pathologist, or lesions of general interest were presented by the chairperson to the PWG for review. The PWG consisted of the quality assessment pathologist and other pathologists experienced in rodent toxicologic pathology. This group examined the tissues without any knowledge of dose groups or previously rendered diagnoses. When the PWG consensus differed from the opinion of the laboratory pathologist, the diagnosis was changed. Thus, the final diagnoses represent a consensus of quality assessment pathologists, the PWG chairperson, and the PWG. Details of these review procedures have been described, in part, by Maronpot and Boorman (1982) and Boorman *et al*. (1985). For subsequent analyses of the pathology data, the diagnosed lesions for each tissue type were evaluated separately or combined according to the guidelines of McConnell *et al*. (1986).

STATISTICAL METHODS

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 found dead of other than natural causes were censored from the survival analyses; animals dying from natural causes were not censored. Statistical analyses for possible dose-related effects on survival used Cox's (1972) method for testing two groups for equality and Tarone's (1975) life table test to identify dose-related trends. All reported P values for the survival analyses are two sided.

Calculation of Incidence

The incidences of neoplasms or nonneoplastic lesions as presented in Tables A1, A5, B1, and B5 are given as the number of animals bearing such lesions at a specific anatomic site and the number of animals with that site examined microscopically. For calculation of statistical significance, the incidences of most neoplasms (Tables A3 and B3) and all nonneoplastic lesions are given as the numbers of animals affected at each site examined microscopically. However, when macroscopic examination was required to detect neoplasms in certain tissues (e.g., harderian gland, intestine, mammary gland, oral cavity, and skin) before microscopic evaluation, or when neoplasms had multiple potential sites of occurrence (e.g., leukemia or lymphoma), the denominators consist of the number of animals on which a necropsy was performed. Tables A3 and B3 also give the survival-adjusted neoplasm rate for each group and each site-specific neoplasm, i.e., the Kaplan-Meier estimate of the neoplasm incidence that would have been observed at the end of the study in the absence of mortality from all other competing risks (Kaplan and Meier, 1958).

Analysis of Neoplasm Incidences

The majority of neoplasms in these studies were considered to be incidental to the cause of death or not rapidly lethal. Thus, the primary statistical method used was logistic regression analysis, which assumed that the diagnosed neoplasms were discovered as the result of death from an unrelated cause and thus did not affect the risk of death. In this approach, neoplasm prevalence was modeled as a logistic function of chemical exposure and time. Both linear and quadratic terms in time were incorporated initially, and the quadratic term was eliminated if the fit of the model was not significantly enhanced. The neoplasm incidences of exposed and control groups were compared on the basis of the likelihood score test for the regression coefficient of dose. This method of adjusting for intercurrent mortality is the prevalence analysis of Dinse and Lagakos (1983), further described and illustrated by Dinse and Haseman (1986). When neoplasms are incidental, this comparison of the time-specific neoplasm prevalences also provides a comparison of the time-specific neoplasm incidences (McKnight and Crowley, 1984).

In addition to logistic regression, other methods of statistical analysis were used, and the results of these tests are summarized in the appendixes. These methods include the life table test (Cox, 1972; Tarone, 1975), appropriate for rapidly lethal neoplasms, and the Fisher exact test and the Cochran-Armitage trend test (Armitage, 1971; Gart *et al*., 1979), procedures based on the overall proportion of neoplasm-bearing animals.

Tests of significance included pairwise comparisons of each exposed group with controls and a test for an overall dose-related trend. Continuity-corrected tests were used in the analysis of neoplasm incidence, and reported P values are one sided. The procedures described in the preceding paragraphs were also used

to evaluate selected nonneoplastic lesions. For further discussion of these statistical methods, refer to Haseman (1984).

Analysis of Nonneoplastic Lesion Incidences

Because all nonneoplastic lesions in this study were considered to be incidental to the cause of death or not rapidly lethal, the primary statistical analysis used was a logistic regression analysis in which nonneoplastic lesion prevalence was modeled as a logistic function of chemical exposure and time. For lesions detected at the interim evaluation, the Fisher exact test, a procedure based on the overall proportion of affected animals, was used.

Analysis of Continuous Variables

Two approaches were employed to assess the significance of pairwise comparisons between exposed and control groups in the analysis of continuous variables. Organ and body weight data, which have approximately normal distributions, were analyzed using the parametric multiple comparison procedures of Dunnett (1955) and Williams (1971, 1972). Hematology data, which have typically skewed distributions, were analyzed using the nonparametric multiple comparison methods of Shirley (1977) and Dunn (1964). Jonckheere's test (Jonckheere, 1954) was used to assess the significance of the dose-related trends and to determine whether a trend-sensitive test (Williams' or Shirley's test) was more appropriate for pairwise comparisons than a test that does not assume a monotonic dose-related trend (Dunnett's or Dunn's test). Prior to analysis, extreme values identified by the outlier test of Dixon and Massey (1951) were examined by NTP personnel, and implausible values were eliminated from the analysis. Average severity values were analyzed for significance using the Mann-Whitney U test (Hollander and Wolfe, 1973).

Reproductive Toxicity Data

Body weight data for F_0 rats, maternal body weight data during gestation and lactation, litter weight data, pup delivery data, percent male pups, and pups surviving on days 4 and 21 were analyzed using Williams' or Dunnett's test. Feed consumption data for F_0 rats were analyzed using Dunn's or Shirley's test.

Historical Control Data

Although the concurrent control group is always the first and most appropriate control group used for evaluation, historical control data can be helpful in the overall assessment of neoplasm incidence in certain instances. Consequently, neoplasm incidences from the NTP historical control database which is updated yearly are included in the NTP reports for neoplasms appearing to show compound-related effects.

QUALITY ASSURANCE METHODS

The reproductive toxicity and 2-year studies were conducted in compliance with Food and Drug Administration Good Laboratory Practice Regulations (21 CFR, Part 58). In addition, as records from the 2-year study were submitted to the NTP Archives, this study was audited retrospectively by an independent quality assurance contractor. Separate audits covering completeness and accuracy of the pathology data, pathology specimens, final pathology tables, and a draft of this NTP Technical Report were conducted. Audit procedures and findings are presented in the reports and are on file at NIEHS. The audit findings were reviewed and assessed by NTP staff, so all comments had been resolved or were otherwise addressed during the preparation of this Technical Report.

GENETIC TOXICOLOGY

The genetic toxicity of D&C Yellow No. 11 was assessed by testing the ability of the chemical to induce mutations in various strains of *Salmonella typhimurium*, sister chromatid exchanges and chromosomal aberrations in cultured Chinese hamster ovary cells, and increases in the frequency of micronucleated erythrocytes in mouse peripheral blood. The protocols for these studies and the results are given in Appendix C.

The genetic toxicity studies of D&C Yellow No. 11 are part of a larger effort by the NTP to develop a database that would permit the evaluation of carcinogenicity in experimental animals from the structure and responses of the chemical in short-term

in vitro and *in vivo* genetic toxicity tests. These genetic toxicity tests were originally developed to study mechanisms of chemically induced DNA damage and to predict carcinogenicity in animals, based on the electrophilic theory of chemical carcinogenesis and the somatic mutation theory (Miller and Miller, 1977; Straus, 1981; Crawford, 1985).

There is a strong correlation between a chemical's potential electrophilicity (structural alert to DNA reactivity), mutagenicity in *Salmonella*, and carcinogenicity in rodents. The combination of electrophilicity and *Salmonella* mutagenicity is highly correlated with the induction of carcinogenicity in rats and mice and/or at multiple tissue sites (Ashby and Tennant, 1991).

Other *in vitro* genetic toxicity tests do not correlate well with rodent carcinogenicity (Tennant *et al*., 1987; Zeiger *et al*., 1990), although these other tests can provide information on the types of DNA and chromosome effects that can be induced by the chemical being investigated. Data from NTP studies show that a positive response in *Salmonella* is currently the most predictive *in vitro* test for rodent carcinogenicity (89% of the *Salmonella* mutagens were rodent carcinogens), and that there is no complementarity among the *in vitro* genetic toxicity tests. That is, no battery of tests that included the *Salmonella* test improved the predictivity of the *Salmonella* test alone. The predictivity for carcinogenicity of a positive response in bone marrow chromosome aberration or micronucleus tests is not yet defined.

TABLE 1
Experimental Design and Materials and Methods in the Feed Studies of D&C Yellow No. 11

Reproductive Toxicity Study	2-Year Study
Study Laboratory	
Southern Research Institute (Birmingham, AL) and Argus Research Laboratories, Inc. (Horsham, PA)	Southern Research Institute (Birmingham, AL)
Strain and Species	
F344/N rats	F344/N rats
Animal Source	
Taconic Farms (Germantown, NY)	Bred in-house
Time Held Before Studies	
10 days	Not applicable
Average Age When Studies Began	
42 days	28 days at weaning
Date of First Dose	
18 December 1989	26 April 1990
Duration of Dosing	
Males: 13 weeks Females: 19 weeks	Males: 105 weeks Females: 106 weeks
Date of Last Dose	
Males: 13 March 1990 Females: 24 April 1990	12-Month interim evaluation — males: 17 April 1991 females: 18 April 1991 Terminal — males: 27 April 1992 females: 4 May 1992
Necropsy Dates	
Not applicable	12-Month interim evaluation — males: 17 April 1991 females: 18 April 1991 Terminal — males: 27-28 April 1992 females: 4-6 May 1992
Average Age at Necropsy	
Not applicable	12-Month interim evaluation — males: 56 weeks females: 56 weeks Terminal — males: 110 weeks females: 111 weeks

TABLE 1
Experimental Design and Materials and Methods in the Feed Studies of D&C Yellow No. 11 (continued)

Reproductive Toxicity Study	2-Year Study
Size of Study Groups 60 males and 60 females	12-Month interim evaluation) 6 to 10 males and 9 to 10 females Terminal) 50 to 54 males and 50 to 51 females
Method of Distribution Rats were distributed randomly into groups of approximately equal initial mean body weights.	Litters culled twice using a table of random numbers to no more than four males and four females per litter on day 4, then two male and two female pups from each litter on day 21; 60 male and 60 female pups per exposure group were continued on study after weaning.
Animals per Cage Before cohabitation: 5 During cohabitation: 1 pair After cohabitation: 5 males or 1 dam and litter	5
Method of Animal Identification Tail tattoo	Tail tattoo
Diet NIH-07 open formula mash (Zeigler Brothers, Inc., Gardners, PA), available *ad libitum*, changed weekly	Same as reproductive toxicity study
Water Distribution Tap water (Birmingham municipal supply) via automatic watering system (Edstrom Industries, Inc., Waterford, WI), available *ad libitum*	Same as reproductive toxicity study
Cages Solid-bottom polycarbonate (Lab Products, Maywood, NJ), changed twice weekly except from day 18 of gestation through delivery	Solid-bottom polycarbonate (Lab Products, Maywood, NJ), changed twice weekly or when excessively soiled or wet
Bedding Sani-Chips (P.J. Murphy Forest Products Corp., Montville, NJ), changed twice weekly except from day 18 of gestation through delivery	Sani-Chips (P.J. Murphy Forest Products Corp., Montville, NJ), changed twice weekly
Rack Filters Reemay® spun-bonded polyester (Andico, Birmingham, AL), changed once every 2 weeks except from day 18 of gestation through delivery	Reemay® spun-bonded polyester (Andico, Birmingham, AL), changed once every 2 weeks
Racks Stainless steel (Lab Products, Inc., Maywood, NJ), changed once every 2 weeks except from day 18 of gestation through delivery	Stainless steel (Lab Products, Inc., Maywood, NJ), changed once every 2 weeks

TABLE 1
Experimental Design and Materials and Methods in the Feed Studies of D&C Yellow No. 11 (continued)

Reproductive Toxicity Study	2-Year Study
Animal Room Environment Temperature: 20.0° to 25.6° C Relative humidity: 23.3% to 81.2% Fluorescent light: 12 hours/day Room air: minimum of 10 changes per hour	Same as reproductive toxicity study
Doses 0, 500, 1,700, or 5,000 ppm in feed, available *ad libitum*	0, 500, 1,700, or 5,000 ppm in feed, available *ad libitum*
Type and Frequency of Observation Observed twice daily; clinical findings and body weights were recorded on day 1 and weekly before cohabitation for F_0 males and females, on days 0, 6, 15, and 21 of gestation for F_0 females, and on days 1, 4, 14, and 21 of lactation for F_0 females and F_1 pups. Feed consumption was recorded by cage weekly before cohabitation, on days 0, 6, 15, and 21 during gestation, and on days 1, 4, 14, and 21 during lactation.	Observed twice daily; animals were weighed and clinical findings were recorded initially, weekly for 13 weeks, monthly thereafter, and at the end of the studies. Feed consumption was recorded during week 2 and approximately monthly thereafter by cage.
Method of Sacrifice CO_2 asphyxiation	CO_2 asphyxiation
Necropsy None	Necropsy performed on all animals. Organs weighed at the 12-month interim evaluation were the liver and right kidney.
Clinical Pathology None	Blood was collected from the retroorbital sinus of all 12-month interim evaluation rats. ***Hematology:*** hematocrit; hemoglobin; erythrocyte, reticulocyte, and nucleated erythrocyte counts; mean cell volume; mean cell hemoglobin; mean cell hemoglobin concentration; platelet counts; and total leukocyte counts and differentials
Histopathology None	Complete histopathology was performed on all rats. In addition to gross lesions and tissue masses, the tissues examined included: adrenal gland, brain, clitoral gland, esophagus, femur with marrow and epiphysis, heart and aorta, large intestine (cecum, colon, and rectum), small intestine (duodenum, jejunum, and ileum), kidneys, liver, lungs and mainstem bronchi, lymph nodes (mandibular and mesenteric), mammary gland, nose, ovaries, pancreas, parathyroid gland, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular stomach), testes, thymus, thyroid gland, trachea, urinary bladder, and uterus.

RESULTS

DOSE SELECTION RATIONALE

The results of the 13-week rat study were used to select doses of 500, 1,700, and 5,000 ppm for the current F344/N rat study. In the 13-week feed study, rats were given 500, 1,700, 5,000, 17,000, or 50,000 ppm. There was no perinatal exposure, and animals were about 6 weeks old when placed on dosed feed. Mean body weights of males and females were significantly reduced after 13 weeks of exposure to 17,000 and 50,000 ppm, and there was mild hepatocellular periportal degeneration in 7 males given 17,000 ppm, in all 10 given 50,000 ppm, and in 2 females given 50,000 ppm. This lesion was minimal at doses of 1,700 and 5,000 ppm in males (4/4, 9/10) and females (2/2, 7/7) and in females at 17,000 ppm (9/10) and was not observed in groups given 500 ppm. In addition, a range-finding study was conducted in which female rats were given 5,000, 17,000, or 50,000 ppm D&C Yellow No. 11 in feed for 4 weeks before mating and during mating, gestation, and the first 4 weeks after having litters. Pups were weaned at week 4 and continued on the same feed as their dams for an additional 4 weeks. Litters would have been potentially exposed *in utero*, through lactation, and feed. There was no difference between study groups in reproductive performance. However, pup body weights in the 17,000 and 50,000 ppm groups were decreased at 8 weeks of age. Microscopic evaluation showed that the liver lesions in exposed pups were similar to those described for the 13-week study.

Following discussions with the FDA, the nominator, the NTP conducted studies of perinatal exposure followed by dietary exposure for 2 years after weaning in male and female F344/N rats to assess the toxicity and carcinogenicity of D&C Yellow No. 11. This study was chosen to generate data similar to those used by the FDA to regulate other color additives, and the results are presented in this Technical Report.

REPRODUCTIVE TOXICITY STUDY

All first-generation (F_0) male and female rats survived until the end of the study. Prior to cohabitation, mean body weight gains of males (days 1 to 71) given 500, 1,700, or 5,000 ppm and of females (days 1 to 66) given 5,000 ppm were significantly lower than those of the controls (Table F1). The mean body weight gains of exposed females during gestation and lactation were generally similar to those of the controls (Table F3). Feed consumption by exposed groups of rats was generally similar to that by the control groups prior to cohabitation (Table F2). Dietary levels of 500, 1,700, and 5,000 ppm D&C Yellow No. 11 resulted in average daily doses of approximately 35, 120, and 350 mg D&C Yellow No. 11/kg body weight to males and 35, 120, and 370 mg/kg to females.

Prior to cohabitation, clinical findings attributed to D&C Yellow No. 11 exposure included yellow discoloration of the entire body or fur in all males and females given 1,700 or 5,000 ppm and in all males and seven females given 500 ppm. All rats given 1,700 or 5,000 ppm had urine-stained abdominal fur. Yellow discoloration of the fur was observed in all exposed female rats during gestation and lactation.

The duration of gestation (Table F3), the average litter size, the number of live pups on days 4 (precull) and 21, and the percent male pups for each exposure group (Table F4) were similar to those of the controls. The mean body weights of exposed litters were significantly less than those of the control litters on days 14 and 21; this effect was considered to be related to D&C Yellow No. 11 exposure.

2-YEAR STUDY

Survival

Estimates of 2-year survival probabilities for second-generation (F_1) male and female rats are shown in Table 2 and in the Kaplan-Meier survival curves (Figure 1). Survival of males given 1,700 or 5,000 ppm was significantly less than that of the controls. Survival of 1,700 ppm females was significantly greater than that of the controls. Survival of 500 ppm males and females and of 5,000 ppm females was similar to that of the controls.

TABLE 2
Survival of Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Male				
Animals initially in study	60	60	60	60
12-Month interim evaluation[a]	10	9	9	6
Moribund	29	29	41	49
Natural deaths	2	2	1	3
Other	0	0	1	0
Animals surviving to study termination	19	20	8	2
Percent probability of survival at end of study[b]	38	39	16	4
Mean survival (days)[c]	625	614	595	567
Survival analysis[d]	P<0.001	P=0.974	P=0.013	P<0.001
Female				
Animals initially in study	60	60	60	60
12-Month interim evaluation[a]	10	9	10	9
Moribund	25	23	12	25
Natural deaths	3	2	1	3
Animals surviving to study termination	22	26	37	23
Percent probability of survival at end of study	44	51	74	45
Mean survival (days)	637	638	654	631
Survival analysis	P=0.882	P=0.769N	P=0.006N	P=1.000N

[a] Censored from survival analyses
[b] Kaplan-Meier determinations based on the number of animals alive on the first day of terminal sacrifice
[c] Mean of all deaths (uncensored, censored, and terminal sacrifice)
[d] The result of the life table trend test (Tarone, 1975) is in the control column, and the results of the life table pairwise comparisons (Cox, 1972) with the controls are in the exposure columns. A lower mortality in an exposure group is indicated by N.

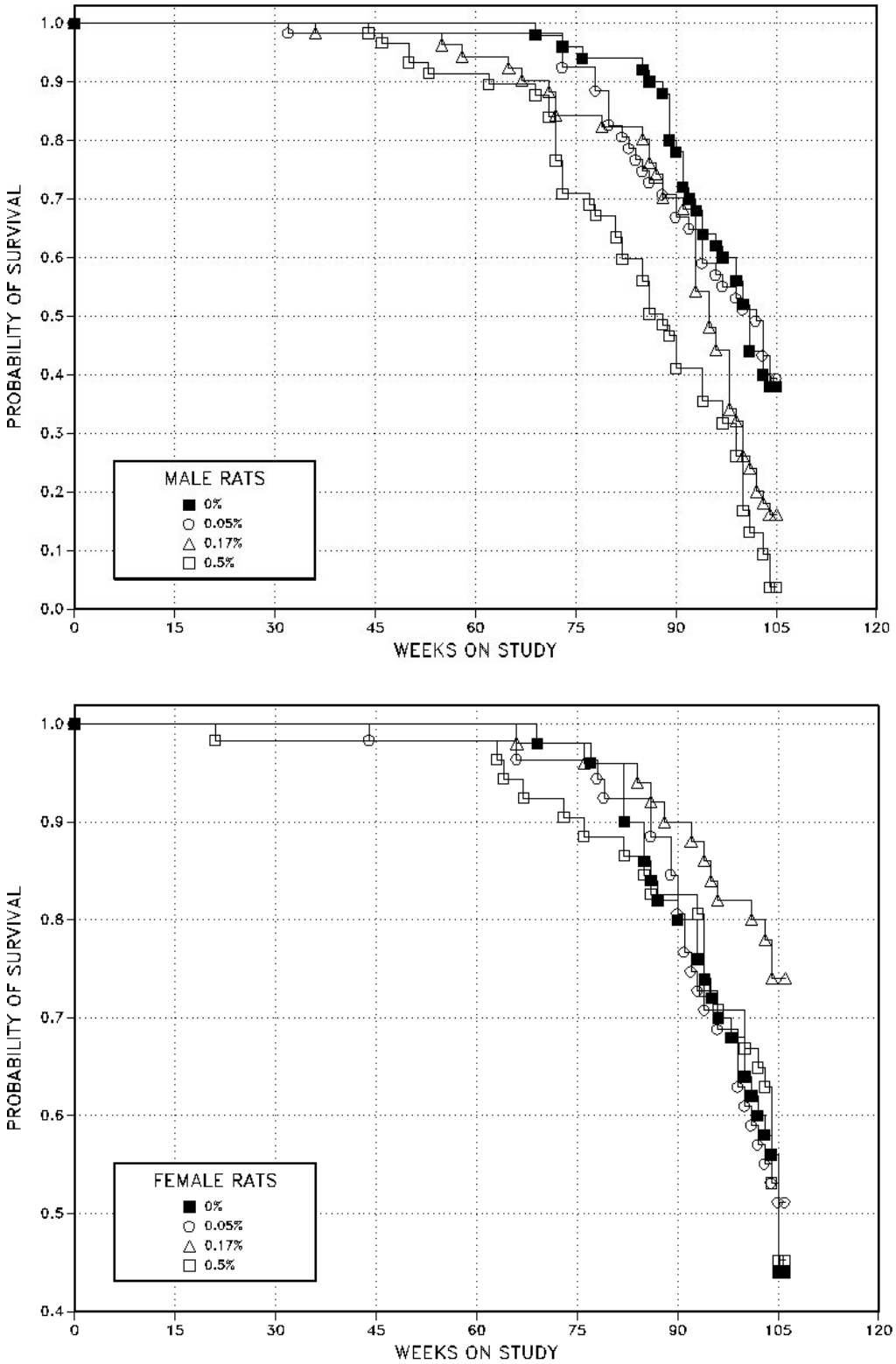


FIGURE 1
Kaplan-Meier Survival Curves for Male and Female Rats Administered D&C Yellow No. 11 in Feed for 2 Years

Body Weights, Feed and Compound Consumption, and Clinical Findings

Male and female F_1 rats were selected from litters in the reproductive toxicity study; therefore, measurements of individual body weight data for F_1 rats in the 2-year study began at weaning when the rats were 28 days old. Mean body weights of 1,700 and 5,000 ppm males and females were generally lower than those of the controls throughout the study (Figure 2; Tables 3 and 4). Final mean body weights of males were 95% (500 ppm), 93% (1,700 ppm), and 85% (5,000 ppm) that of the controls, and those of females were 99%, 95%, and 94% that of the controls. Feed consumption by exposed groups was similar to that by the controls (Tables H1 and H2). Dietary levels of 500, 1,700, and 5,000 ppm D&C Yellow No. 11 resulted in average daily doses of approximately 25, 85, and 250 mg D&C Yellow No. 11/kg body weight to males and 25, 100, and 280 mg/kg to females. Chemical-related clinical findings included yellow discoloration of the entire body in all exposed males and females from day 1 and head swelling and edema in 1,700 and 5,000 ppm males. One 1,700 ppm male and five 5,000 ppm males were killed moribund between weeks 49 and 81; these deaths were attributed to extensive edema.

Hematology

A few minimal hematology differences occurred in male rats at the 12-month interim evaluation (Table E1). There was evidence of minimal anemia in exposed males; this anemia was characterized by decreased hematocrit values, hemoglobin concentrations, and erythrocyte counts. There were no differences in the mean cell volume or mean cell hemoglobin concentration in exposed rats, to indicate that erythrocytes were normocytic and normochromic. There were no increases in reticulocyte counts to indicate a bone marrow response to the anemia. Therefore, the minimal anemia was characterized as normocytic, normochromic, and nonresponsive. Normocytic, normochromic, nonresponsive anemias have been related to selective suppression of erythropoiesis in a variety of disorders and may be due to decreased erythropoietin elaboration, bone marrow suppression, or defective iron metabolism. There were no biologically or statistically significant differences in hematology parameters between control and exposed females.

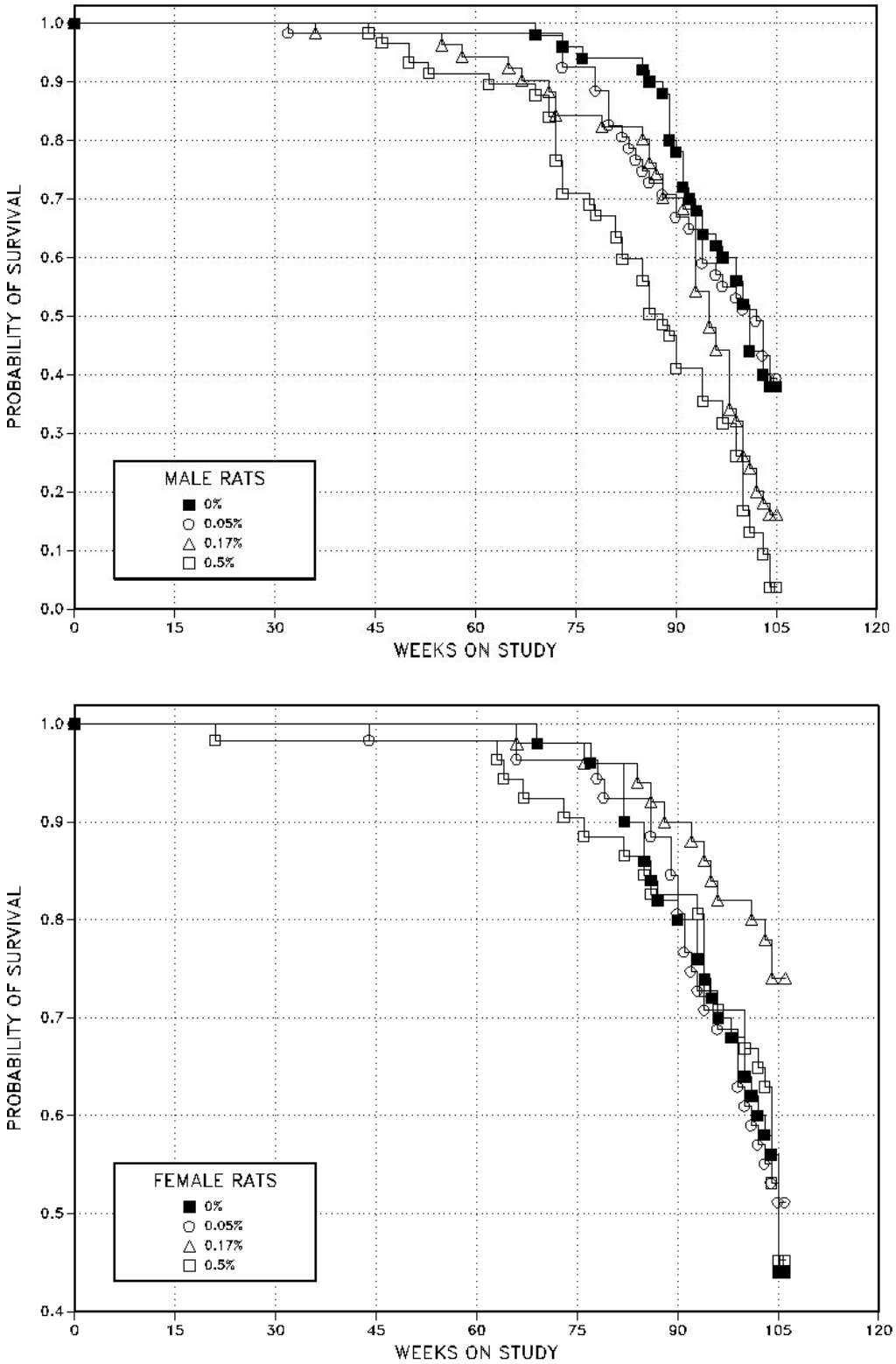


FIGURE 1
Kaplan-Meier Survival Curves for Male and Female Rats Administered D&C Yellow No. 11 in Feed for 2 Years

TABLE 3
Mean Body Weights and Survival of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11

Weeks on Study	0 ppm		500 ppm			1,700 ppm			5,000 ppm		
	Av. Wt. (g)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors
1	112	60	100	89	60	99	88	60	93	83	60
2	159	60	145	91	60	142	89	60	133	84	60
3	194	60	180	93	60	176	90	60	167	86	60
4	227	60	209	92	60	204	90	60	197	87	60
5	254	60	235	92	60	229	90	60	221	87	60
6	277	60	260	94	60	253	91	60	240	87	60
7	288	60	272	94	60	267	93	60	256	89	60
8	303	60	289	96	60	284	94	60	271	90	60
9	315	60	302	96	60	296	94	60	284	90	60
10	327	60	315	96	60	310	95	60	297	91	60
11	337	60	324	96	60	319	95	60	307	91	60
12	347	60	336	97	60	331	96	60	317	91	60
13	353	60	343	97	60	337	96	60	323	91	60
17	383	60	371	97	60	365	96	60	352	92	60
21	401	60	390	97	60	385	96	60	370	93	60
25	416	60	405	97	60	400	96	60	384	92	60
29	431	60	421	98	60	413	96	60	400	93	60
33	425	60[a]	427	100	59[a]	423	100	60	404	95	60[a]
37	449	60	436	97	59	432	96	59	416	93	60
41	455	60	443	97	59	439	97	59	421	93	60
45	456	60	449	98	59	443	97	59	423	93	59
49	464	60	454	98	59	450	97	59	430	93	58
53[b]	472	50	460	98	50	457	97	50	438	93	50
57	472	50	461	98	50	460	97	49	438	93	49
61	475	50	462	97	50	461	97	48	441	93	49
65	477	50	463	97	50	460	96	47	439	92	48
69	475	50	463	97	50	462	97	46	438	92	47
73	472	48	460	98	48	456	97	43	436	93	39
77	473	47	451	96	47	451	95	43	433	92	37
81	470	47	446	95	42	442	94	42	425	91	35
85	463	46	445	96	38	435	94	41	413	89	30
89	452	42	439	97	36	433	96	36	404	89	25
93	451	35	425	94	33	422	94	30	401	89	22
97	446	30	425	95	28	420	94	23	387	87	19
101	435	24	414	95	26	403	93	13	368	85	8
Mean for weeks											
1-13	269		255	95		250	93		239	89	
14-52	431		422	98		417	97		400	93	
53-101	464		447	96		443	95		420	91	

[a] The number of animals weighed for this week is less than the number of animals surviving.
[b] Interim evaluation occurred during week 51.

TABLE 4
Mean Body Weights and Survival of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11

Weeks on Study	0 ppm		500 ppm			1,700 ppm			5,000 ppm		
	Av. Wt. (g)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors
1	100	60	89	90	60	88	89	60	85	86	60
2	126	60	117	93	60	115	92	60	113	90	60
3	141	60	132	93	60	130	92	60	129	92	60
4	152	60	145	95	60	143	94	60	143	94	60
5	163	60	154	95	60	150	92	60	151	93	60[a]
6	168	60	161	96	60	156	93	60	155	92	60
7	177	60	168	95	60	164	93	60	163	92	60
8	180	60	173	96	60	167	93	60	168	93	60
9	186	60	178	96	60	174	93	60	173	93	60
10	187	60	179	96	60	173	92	60	174	93	60
11	193	60	185	96	60	181	94	60	180	93	60
12	196	60	189	96	60	183	93	60	184	93	60
16	206	60	200	97	60	196	95	60	194	94	60
21	212	60	205	97	60	201	95	60	197	93	60
24	217	60	212	97	60	207	95	60	205	94	59
28	227	60	218	96	60	215	95	60	211	93	59
32	233	60	227	97	60	221	95	60	219	94	59
36	238	60	231	97	60	222	93	60	223	94	59
40	246	60	240	98	60	231	94	60	233	95	59
44	255	60	248	97	59	238	93	60	240	94	59
48	265	60	258	97	59	251	95	60	250	94	59
52[b]	277	50	276	100	50	259	94	50	263	95	50
56	290	50	285	99	50	273	94	50	275	95	50
60	296	50	293	99	50	280	95	50	282	95	50
64	307	50	303	99	50	289	94	50	291	95	48
68	314	50	312	99	49	300	96	49	299	95	47
72	320	49	317	99	49	304	95	49	307	96	47
76	323	49	320	99	49	305	95	49	308	95	45
80	331	48	323	98	47	310	94	48	313	95	45
84	339	45	330	97	47	315	93	47	317	93	44
88	342	41	330	97	45	320	94	45	319	93	42
92	348	40	333	96	38	327	94	44	320	92	42
96	357	35	342	96	35	331	93	41	332	93	36
100	355	33	344	97	31	336	95	41	334	94	35
104	354	28	349	99	27	337	95	37	333	94	27
Mean for weeks											
1-13	164		156	95		152	93		152	93	
14-52	238		232	97		224	94		224	94	
53-104	329		322	98		310	94		310	94	

[a] The number of animals weighed for this week is less than the number of animals surviving.
[b] Interim evaluation occurred during week 52.

Pathology and Statistical Analyses

This section describes the statistically significant or biologically noteworthy changes in the incidences of mononuclear cell leukemia and neoplasms and/or nonneoplastic lesions of the liver, kidney, oral cavity (oral mucosa and tongue), testis, forestomach, small intestine, salivary gland, pancreas, lymph nodes, clitoral gland, and pituitary gland. Summaries of the incidences of neoplasms and nonneoplastic lesions, individual animal tumor diagnoses, statistical analyses of primary neoplasms that occurred with an incidence of at least 5% in at least one animal group, and historical incidences for the neoplasms mentioned in this section are presented in Appendix A for male rats and Appendix B for female rats.

Liver: At the 12-month interim evaluation, absolute and relative liver weights of all exposed groups of males and females were significantly greater than those of the controls (Table D1). At 2 years, the incidences of hepatocellular adenoma in 5,000 ppm males and of hepatocellular adenoma or carcinoma (combined) in 5,000 ppm females were significantly greater than those in the controls (Tables 5, A3, and B3); these neoplasms occurred with significant exposure-related trends. The incidence of hepatocellular adenoma in 5,000 ppm males exceeded the historical range (0% to 10%; Tables 5 and A4a) in untreated controls from NTP feed studies; the incidences of adenoma or carcinoma (combined) in 1,700 and 5,000 ppm females also exceeded the historical control range (0% to 6%; Tables 5 and B4a). Hepatocellular adenomas were discrete masses with distinct borders that compressed and replaced adjacent hepatic parenchyma (Plate 1). Hepatic cords within adenomas typically were at sharp angles to the cords in the adjacent normal hepatic parenchyma. Adenomas had loss of normal lobular pattern and usually lacked central veins and portal areas. Cells within adenomas were often somewhat pleomorphic and had altered staining patterns. The hepatocellular carcinoma (Plate 2) in a 5,000 ppm female was a discrete lesion with markedly disturbed architecture (clumps of cells separated by irregular, relatively wide spaces) and more cellular atypia than the adenomas.

At 12 months, the incidences of clear cell foci in 1,700 and 5,000 ppm females were significantly greater than that in the controls (Tables 5 and B5). At 2 years, the incidences of mixed cell foci in exposed males and of clear cell foci in exposed males (except 500 ppm) and females were significantly greater than those in the controls (Tables 5, A5, and B5). In 1,700 ppm males at 2 years, the incidence of eosinophilic foci was significantly greater than that of the controls. At 12 months and 2 years, the incidences of basophilic foci in 1,700 and 5,000 ppm females appeared to be significantly less than those of the controls; however, basophilic foci may have been obscured by cytologic alterations. Foci of hepatocellular alteration were discrete areas within the liver with a relatively normal lobular architecture but having altered staining characteristics (Plate 3).

The incidences of cytologic alterations of hepatocytes in all exposed groups of males and females were significantly greater than those in the controls at 12 months and 2 years (Tables 5, A5, and B5), and the severities generally increased with increasing exposure concentration. Cytologic alterations of hepatocytes consisted of increases in basophilia and granularity in the cytoplasm of hepatocytes that involved primarily periportal hepatocytes in mildly affected cases while more severely affected livers had diffuse involvement. The increased basophilia and granularity of the cytoplasm of hepatocytes in many exposed rats probably obscured detection of basophilic foci. The incidences of bile duct pigmentation in all exposed groups of males and females at 12 months and 2 years, of hepatocyte pigmentation in exposed males and females at 12 months (except 500 ppm males) and 2 years, and of Kupffer cell pigmentation in 5,000 ppm males and females at 12 months and in 1,700 and 5,000 ppm males and females at 2 years were significantly greater than those in the controls. The severities of bile duct pigmentation and hepatocyte pigmentation generally increased with increasing exposure concentration. Pigmentation was a minimal to moderate accumulation of a golden to green-brown granular material within the cytoplasm of hepatocytes, bile duct epithelium, or, less commonly, Kupffer cells (Plate 4). Special stains of pigment in the 14-day study (NTP, 1991a) were negative for hemosiderin, bile, and lipofuscin. The incidences of bile duct hyperplasia in 1,700 and 5,000 ppm females at 12 months and 2 years were significantly greater than in the controls; however, the incidences in exposed males were significantly less than in the controls at 2 years.

TABLE 5
Incidences of Neoplasms and Nonneoplastic Lesions of the Liver in Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Male				
12-Month Interim Evaluation				
Number Examined Microscopically	10	9	9	6
Basophilic Focus[a]	1	1	0	1
Clear Cell Focus	0	0	1	1
Eosinophilic Focus	0	2	0	0
Mixed Cell Focus	0	0	0	1
Bile Duct, Hyperplasia	4 (1.3)[b]	0	0	1 (1.0)
Bile Duct, Pigmentation	0	9** (1.0)	9** (1.0)	6** (1.3)
Hepatocyte, Cytologic Alterations	0	8** (1.0)	9** (1.6)	6** (1.7)
Hepatocyte, Pigmentation	0	1 (1.0)	8** (1.0)	6** (1.2)
Kupffer Cell, Pigmentation	0	0	0	5** (1.0)
2-Year Study				
Number Examined Microscopically	50	51	51	54
Basophilic Focus	14	8	6	7
Clear Cell Focus	9	15	15*	18**
Eosinophilic Focus	7	5	14*	12
Mixed Cell Focus	1	10**	9**	10**
Bile Duct, Hyperplasia	49 (2.1)	26** (1.5)	18** (1.4)	32** (1.5)
Bile Duct, Pigmentation	0	38** (1.2)	51** (1.9)	54** (2.2)
Hepatocyte, Cytologic Alterations	0	20** (2.1)	44** (2.4)	42** (2.7)
Hepatocyte, Pigmentation	0	22** (1.0)	45** (1.7)	51** (2.1)
Kupffer Cell, Pigmentation	7 (2.4)	15 (2.0)	23** (1.9)	26** (1.8)
Hepatocellular Adenoma[c]				
Overall rate[d]	1/50 (2%)	2/51 (4%)	1/51 (2%)	7/54 (13%)
Adjusted rate[e]	5.3%	7.9%	2.6%	75.0%
Terminal rate[f]	1/19 (5%)	1/20 (5%)	0/8 (0%)	1/2 (50%)
First incidence (days)	733 (T)	656	607	498
Logistic regression test[g]	P=0.001	P=0.487	P=0.757	P=0.008
Female				
12-Month Interim Evaluation				
Number Examined Microscopically	10	9	10	9
Basophilic Focus	7	2	2*	1*
Clear Cell Focus	0	1	4*	4*
Eosinophilic Focus	0	0	1	0
Bile Duct, Hyperplasia	1 (1.0)	1 (1.0)	6* (1.0)	9** (1.4)
Bile Duct, Pigmentation	0	9** (1.2)	7** (1.0)	9** (2.3)
Hepatocyte, Cytologic Alterations	0	4* (1.0)	10** (1.5)	9** (2.1)
Hepatocyte, Pigmentation	0	9** (1.0)	10** (1.3)	9** (2.6)
Kupffer Cell, Pigmentation	0	0	0	9** (1.4)

(continued)

TABLE 5
Incidences of Neoplasms and Nonneoplastic Lesions of the Liver in Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Female (continued)				
2-Year Study				
Number Examined Microscopically	50	51	50	51
Basophilic Focus	32	26	11**	12**
Clear Cell Focus	10	18*	29**	30**
Eosinophilic Focus	10	9	14	16
Mixed Cell Focus	12	19	20	16
Bile Duct, Hyperplasia	14 (1.3)	10 (1.7)	27** (1.5)	33** (1.5)
Bile Duct, Pigmentation	0	46** (1.3)	49** (2.0)	50** (2.4)
Hepatocyte, Cytologic Alterations	0	11** (2.3)	31** (2.1)	40** (2.5)
Hepatocyte, Pigmentation	0	34** (1.1)	44** (2.1)	50** (2.4)
Kupffer Cell, Pigmentation	9 (2.1)	11 (2.4)	16* (1.8)	32** (1.9)
Hepatocellular Adenoma				
Overall rate	0/50 (0%)	2/51 (4%)	5/50 (10%)	4/51 (8%)
Adjusted rate	0.0%	6.4%	13.5%	15.7%
Terminal rate	0/22 (0%)	1/26 (4%)	5/37 (14%)	3/23 (13%)
First incidence (days)	—[h]	645	740 (T)	720
Logistic regression test	P=0.100	P=0.241	P=0.095	P=0.068
Hepatocellular Carcinoma				
Overall rate	0/50 (0%)	0/51 (0%)	0/50 (0%)	1/51 (2%)
Hepatocellular Adenoma or Carcinoma[i]				
Overall rate	0/50 (0%)	2/51 (4%)	5/50 (10%)	5/51 (10%)
Adjusted rate	0.0%	6.4%	13.5%	18.5%
Terminal rate	0/22 (0%)	1/26 (4%)	5/37 (14%)	3/23 (13%)
First incidence (days)	—	645	740 (T)	720
Logistic regression test	P=0.042	P=0.241	P=0.095	P=0.036

* Significantly different ($P \leq 0.05$) from the control group by the Fisher exact test (interim evaluation) or the logistic regression test (2-year study)
** $P \leq 0.01$
(T) Terminal sacrifice
[a] Number of animals with lesion
[b] Average severity of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked
[c] Historical incidence for 2-year NTP feed studies with untreated controls (mean ± standard deviation): 30/1,301 (2.3% ± 2.9%); range, 0%-10%
[d] Number of animals with neoplasm per number of animals with liver examined microscopically
[e] Kaplan-Meier estimated neoplasm incidence at the end of the study after adjustment for intercurrent mortality
[f] Observed incidence in animals surviving until the end of the study
[g] In the control column are the P values associated with the trend test. In the exposed group columns are the P values corresponding to the pairwise comparisons between the controls and that exposed group. The logistic regression test regards lesions in animals dying prior to terminal kill as nonfatal.
[h] Not applicable; no neoplasms in animal group
[i] Historical incidence: 9/1,301 (0.7% ± 1.5%); range, 0%-6%

Kidney: Two renal tubule adenomas in 5,000 ppm males and one renal tubule carcinoma in a 1,700 ppm male were observed in the standard (single section) evaluation (Tables 6 and A1). Because of this suggestion of a chemical-related increase in renal tubule neoplasms in males, an extended evaluation (step sections) of the kidney was conducted. During the extended evaluation, two additional renal tubule adenomas were observed in 5,000 ppm males, four renal tubule adenomas were observed in 1,700 ppm males, and two renal tubule adenomas were observed in 500 ppm males. No renal tubule neoplasms were observed in male controls. Renal tubule adenomas (Plate 5) were more than five times the diameter of a normal tubule, usually had more complex structures than hyperplasias, and often consisted of clusters of multiple tubule-like structures. The one renal tubule carcinoma in a 1,700 ppm male was approximately 0.5 cm in diameter and was composed of atypical epithelial cells forming solid clusters or abnormal tubule-like structures that invaded the adjacent renal parenchyma. One renal tubule carcinoma also occurred in a 1,700 ppm female (Tables 6 and B1). Renal tubule hyperplasia was observed in exposed groups of males but not in controls, and the incidences in 1,700 ppm males from both standard and extended evaluations were significantly greater than those in the controls (Table 6). Renal tubule hyperplasia was a discrete lesion ranging from a solid cluster of epithelial cells two to three times the diameter of a normal tubule to a cystic lesion consisting of a tubule dilated up to five times the normal diameter and lined with multiple layers of epithelial cells.

At 12 months and 2 years, nephropathy was observed in all control and exposed male rats and in most female rats (Tables 6, A5, and B5). The severity of nephropathy in exposed males and females was significantly greater than in the controls, and the severity was greater in males than in females. Nephropathy included necrosis and regeneration of renal tubule epithelium, typically with increased thickness of basement membrane around regenerative tubules; dilated tubules usually containing proteinaceous fluid; and interstitial fibrosis and inflammatory cell aggregates. At 2 years, the incidences of hyperplasia of transitional epithelium of the kidney, which commonly accompanies advanced nephropathy, were greater in exposed males and 1,700 ppm females than in the controls, and the severity of this lesion in exposed males and females was greater than in the controls. Increased incidences of hyperplasia of the parathyroid gland (0 ppm, 3/47; 500 ppm, 9/47; 1,700 ppm, 15/48; 5,000 ppm, 17/52) and fibrous osteodystrophy of the bone (2/50, 8/51, 18/51, 14/54) in exposed males at 2 years (Table A5) were probably secondary to the impaired kidney function associated with nephropathy.

The incidences of renal tubule pigmentation in all exposed groups of males and females at 12 months and 2 years were significantly greater than those in the controls (Tables 6, A5, and B5). Pigmentation of the renal tubule epithelium was yellow to brown granular material within the cytoplasm of cortical tubule epithelial cells (Plate 6) and was similar in appearance to that seen in the liver.

TABLE 6
Incidences of Neoplasms and Nonneoplastic Lesions of the Kidney in Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Male				
12-Month Interim Evaluation				
Number Examined Microscopically	10	9	9	6
Nephropathy[a]	10 (1.1)[b]	9 (1.9)**	9 (2.6)**	6 (2.3)**
Renal Tubule, Pigmentation	0	7** (1.0)	9** (1.8)	6** (2.5)
2-Year Study				
Single Sections (Standard Evaluation)				
Number Examined Microscopically	50	51	51	54
Nephropathy	50 (2.3)	51 (2.8)**	51 (3.2)**	54 (3.0)**
Renal Tubule, Hyperplasia	0	0	4* (2.0)	3 (2.7)
Renal Tubule, Pigmentation	18 (2.1)	43** (1.8)	47** (2.3)	54** (2.5)
Transitional Epithelium, Hyperplasia	11 (1.3)	23** (1.8)	29** (1.9)	34** (1.7)
Renal Tubule Adenoma[c]				
Overall rate[d]	0/50 (0%)	0/51 (0%)	0/51 (0%)	2/54 (4%)
Renal Tubule Carcinoma				
Overall rate	0/50 (0%)	0/51 (0%)	1/51 (2%)	0/54 (0%)
Step Sections (Extended Evaluation)				
Number Examined Microscopically	50	51	51	54
Renal Tubule, Hyperplasia	0	2 (1.5)	9** (1.6)	2 (1.0)
Renal Tubule Adenoma				
Overall rate	0/50 (0%)	2/51 (4%)	4/51 (8%)	2/54 (4%)
Adjusted rate[e]	0.0%	7.1%	22.5%	18.8%
Terminal rate[f]	0/19 (0%)	1/20 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	—[h]	558	649	678
Logistic regression test[g]	P=0.259	P=0.255	P=0.046	P=0.120
Single Sections and Step Sections (Combined)				
Number Examined Microscopically	50	51	51	54
Renal Tubule, Hyperplasia	0	2	13**	4*
Renal Tubule Adenoma				
Overall rate	0/50 (0%)	2/51 (4%)	4/51 (8%)	4/54 (7%)
Adjusted rate	0.0%	7.1%	22.5%	38.3%
Terminal rate	0/19 (0%)	1/20 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	—	558	649	658
Logistic regression test	P=0.032	P=0.255	P=0.046	P=0.014
Renal Tubule Carcinoma				
Overall rate	0/50 (0%)	0/51 (0%)	1/51 (2%)	0/54 (0%)
Renal Tubule Adenoma or Carcinoma				
Overall rate	0/50 (0%)	2/51 (4%)	5/51 (10%)	4/54 (7%)
Adjusted rate	0.0%	7.1%	26.1%	38.3%
Terminal rate	0/19 (0%)	1/20 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	—	558	649	658
Logistic regression test	P=0.036	P=0.255	P=0.022	P=0.014

(continued)

TABLE 6
Incidences of Neoplasms and Nonneoplastic Lesions of the Kidney in Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Female				
12-Month Interim Evaluation				
Number Examined Microscopically	10	9	10	9
Nephropathy	6 (1.0)	6 (1.0)	7 (1.1)	9 (1.2)
Renal Tubule, Pigmentation	0	9** (1.7)	10** (2.2)	9** (3.0)
Transitional Epithelium, Hyperplasia	0	0	0	2 (1.5)
2-Year Study				
Number Examined Microscopically	50	51	50	51
Nephropathy	45 (1.4)	47 (1.7)*	46 (1.8)**	50* (2.1)**
Renal Tubule, Pigmentation	10 (1.3)	48** (1.8)	50** (2.8)	51** (3.2)
Transitional Epithelium, Hyperplasia	2 (1.0)	6 (1.3)	10* (1.5)	3 (2.3)
Renal Tubule Carcinoma[i]				
Overall rate	0/50 (0%)	0/51 (0%)	1/50 (2%)	0/51 (0%)

* Significantly different ($P \leq 0.05$) from the control group by the Fisher exact test (incidences at interim evaluation), the logistic regression test (incidences at 2 years), or the Mann-Whitney U test (severity of nephropathy)
** $P \leq 0.01$
[a] Number of animals with lesion
[b] Average severity of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked
[c] Historical incidence for 2-year NTP feed studies with untreated controls (mean ± deviation): 9/1,301 (0.7% ± 1.5%); range, 0%-6%
[d] Number of animals with neoplasm per number of animals with kidney examined microscopically
[e] Kaplan-Meier estimated neoplasm incidence at the end of the study after adjustment for intercurrent mortality
[f] Observed incidence in animals surviving until the end of the study
[g] In the control column are the P values associated with the trend test. In the exposed group columns are the P values corresponding to the pairwise comparisons between the controls and that exposed group. The logistic regression test regards lesions in animals dying prior to terminal kill as nonfatal.
[h] Not applicable; no neoplasms in animal group
[i] Historical incidence: 1/1,298 (0.1% ± 0.4%); range, 0%-2%

Oral Cavity (Oral Mucosa and Tongue): Squamous cell carcinomas of the tongue were observed in one 500 ppm male at 12 months and one 5,000 ppm female at 2 years (Tables 7, A1, and B1). One squamous cell carcinoma of the oral mucosa was observed in each group of exposed males and in one female in the 5,000 ppm group at 2 years. Observations of squamous cell carcinoma of the oral cavity in male F344/N rats are extraordinarily unusual because this lesion has never been observed in 1,304 historical control males from NTP feed studies (Table A4c). The incidence for 5,000 ppm female rats also exceeded the historical control range (Table B4c). Squamous cell carcinoma was an irregular mass composed of thick cords and solid clusters of atypical epithelial cells that invaded the underlying connective tissue (Plate 7). At 2 years, squamous cell papillomas were observed in the oral cavity (oral mucosa or tongue) in one control, one 500 ppm, two 1,700 ppm, and four 5,000 ppm males; this lesion was also observed in one control and one 500 ppm female (Tables 7 and B1). The incidence of squamous cell papilloma or squamous cell carcinoma (combined) in 1,700 and 5,000 ppm males exceeds the NTP historical control range (0% to 4%, Table A4c). Squamous cell papilloma was a discrete mass of thick, branching epithelium overlying a central connective tissue core with a stalk-like connection to the mucosal surface (Plate 8). Hyperplasia was identified in the oral mucosa of one 5,000 ppm male at the 12-month interim evaluation and in the tongue of two 5,000 ppm males at 2 years. Hyperplasia was characterized by an increased number of cell layers of mucosal epithelium.

TABLE 7
Incidences of Neoplasms and Nonneoplastic Lesions of the Oral Cavity in Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Male				
12-Month Interim Evaluation				
Oral Mucosa[a]	0	0	0	1
Hyperplasia[b]	0	0	0	1 (2.0)[c]
Tongue	0	1	0	0
Squamous Cell Carcinoma	0	1	0	0
2-Year Study				
Tongue	1	0	1	4
Hyperplasia	0	0	0	2 (2.5)
Oral Cavity (Oral Mucosa or Tongue)				
Squamous Cell Papilloma				
Overall rate[d]	1/50 (2%)	1/51 (2%)	2/51 (4%)	4/54 (7%)
Adjusted rate[e]	5.3%	3.2%	6.5%	28.1%
Terminal rate[f]	1/19 (5%)	0/20 (0%)	0/8 (0%)	0/2 (0%)
First incidence (days)	733 (T)	658	406	481
Logistic regression test[g]	P=0.087	P=0.755	P=0.606	P=0.110
Squamous Cell Carcinoma[h]				
Overall rate	0/50 (0%)	1/51(2%)	1/51 (2%)	1/54 (2%)
Squamous Cell Papilloma or Squamous Cell Carcinoma[i]				
Overall rate	1/50 (2%)	2/51 (4%)	3/51 (6%)	5/54 (9%)
Adjusted rate	5.3%	6.9%	10.6%	30.4%
Terminal rate	1/19 (5%)	0/20 (0%)	0/8 (0%)	0/2 (0%)
First incidence (days)	733 (T)	658	406	481
Logistic regression test	P=0.066	P=0.487	P=0.369	P=0.069

(continued)

TABLE 7
Incidences of Neoplasms and Nonneoplastic Lesions of the Oral Cavity in Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Female				
2-Year Study				
Oral Cavity (Oral Mucosa or Tongue)				
Squamous Cell Papilloma				
Overall rate	1/50 (2%)	1/51 (2%)	0/50 (0%)	0/51 (0%)
Squamous Cell Carcinoma[j]				
Overall rate	0/50 (0%)	0/51 (0%)	0/50 (0%)	2/51 (4%)
Squamous Cell Papilloma or Squamous Cell Carcinoma[k]				
Overall rate	1/50 (2%)	1/51 (2%)	0/50 (0%)	2/51 (4%)
Adjusted rate	4.5%	2.3%	0.0%	8.2%
Terminal rate	1/22 (5%)	0/26 (0%)	0/37 (0%)	1/23 (4%)
First incidence (days)	740 (T)	628	)[l]	733
Logistic regression test	P=0.332	P=0.757N	P=0.396N	P=0.518

(T)Terminal sacrifice
[a] Number of animals with tissue examined microscopically
[b] Number of animals with lesion
[c] Average severity of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked
[d] Number of animals with neoplasm per number of animals necropsied
[e] Kaplan-Meier estimated neoplasm incidence at the end of the study after adjustment for intercurrent mortality
[f] Observed incidence in animals surviving until the end of the study
[g] In the control column are the P values associated with the trend test. In the exposed group columns are the P values corresponding to the pairwise comparison between the controls and that exposed group. The logistic regression test regards lesions in animals dying prior to terminal kill as nonfatal. A lower incidence in an exposure group is indicated by N.
[h] Historical incidence for 2-year NTP feed studies with untreated control groups: 0/1,304
[i] Historical incidence (mean ± standard deviation): 10/1,304 (0.8% ± 1.3%); range, 0%-4%
[j] Historical incidence: 4/1,301 (0.3% ± 0.7%); range, 0%-2%
[k] Historical incidence: 15/1,301 (1.2% ± 1.6%); range, 0%-6%
[l] Not applicable; no neoplasms in animal group

Testis: There was an exposure-related increase in the incidences of testicular adenoma at 2 years, and the incidence of this lesion in each exposed group was significantly greater than that in the controls (0 ppm, 39/49; 500 ppm, 46/51; 1,700, 48/51; 5,000 ppm, 46/54; Table A3). Because the incidences within the historical control range (74% to 98%; Table A4d) are so high, the significance of the incidences in the exposed groups is unclear.

Forestomach and Small Intestine: The incidences of mucosal hyperplasia of the forestomach in exposed males (4/50, 13/51, 19/51, 21/54; Table A5) and females (5/50, 17/51, 30/50, 27/51; Table B5) were greater than those in the controls at 2 years. Hyperplasia of the squamous epithelium of the forestomach varied in severity and extent, ranging from minimal focal lesions at the limiting ridge of the mucosa to marked lesions affecting cell layers throughout the forestomach mucosa. In the small intestine of 1,700 and 5,000 ppm males at 2 years, the incidences of epithelial hyperplasia of the duodenum (1/50, 4/51, 22/51, 21/54), jejunum (0/50, 3/50, 10/51, 12/54), and ileum (0/49, 3/50, 10/50, 8/54) were greater than those in the controls. Hyperplasia of the small intestine consisted of a diffuse increase in the number of villous epithelial cells, which appeared crowded together and taller, and an increase in the height of the villous projections. This lesion was usually apparent on gross examination because of the greater diameter and thicker mucosa of affected intestines.

Salivary Gland and Pancreas: Incidences of atrophy of the salivary glands were greater than those in the controls in 5,000 ppm males (1/50, 1/51, 5/50, 7/54; Table A5) and in all exposed groups of females (0/50, 8/46, 7/50, 13/50; Table B5). Atrophy was a minimal to mild focal to multifocal decrease in the size of glandular acini accompanied by increased amounts of interstitial connective tissue between acini. In the pancreas, incidences of cytoplasmic alteration of the acinar cell in exposed males were significantly greater than that in the controls (0/50, 5/51, 11/51, 8/54; Table A5). Cytoplasmic alteration of the acinar cells of the pancreas was a diffuse loss of zymogen granules from the cytoplasm. This change might reflect debilitation because all males in which this change occurred died before the end of the study. The incidences (9/50, 19/51, 17/50, 14/51; Table B5), but not the severity, of pancreatic atrophy were greater in exposed females than in controls. Pancreatic atrophy was characterized by a decrease in the size of pancreatic acini and a relative increase in the amount of connective tissue between acini.

Lymph Nodes: At 2 years, the incidences of lymphoid hyperplasia were greater than those in the controls in the mandibular lymph nodes in 1,700 and 5,000 ppm males (8/50, 12/51, 22/50, 25/53) and females (5/50, 10/51, 14/50, 13/49), the mesenteric lymph nodes in 1,700 and 5,000 ppm males (3/50, 2/50, 10/51, 14/54), and the mediastinal lymph nodes in 1,700 and 5,000 ppm males (0/20, 0/23, 9/26, 17/45) (Tables A5 and B5). Mediastinal and pancreatic lymph nodes were examined microscopically only when they were grossly abnormal. Lymphoid hyperplasia was described as an increase in the size (1.5 to 2 times normal) of lymph nodes, which was usually accompanied by an increase in the density of cortical lymphocytes. This lymphoid hyperplasia suggests an immune response was associated with the administration of D&C Yellow No. 11 in some individuals. In 1,700 and 5,000 ppm males, the incidences of hemorrhage of the mesenteric lymph nodes (0/50, 0/50, 9/51, 7/54) and mediastinal lymph nodes (0/20, 3/23, 7/26, 16/45) were greater than those in the controls. Hemorrhage within lymph nodes consisted of small to moderate numbers of extravascular red blood cells within medullary sinuses. Incidences of pigmentation of the pancreatic lymph nodes in 5,000 ppm males (1/20, 3/23, 3/26, 12/45) and females (1/9, 2/11, 6/11, 8/15) were greater than in the controls. Pigmentation within lymph nodes was described as yellow to brown granular material within the cytoplasm of macrophages.

Clitoral Gland: At 2 years, the incidences of clitoral gland adenoma (11/49, 4/50, 5/49, 4/51) and clitoral gland adenoma or carcinoma (combined) (17/49, 6/50, 11/49, 6/51) in exposed groups of females were significantly less than those in the controls (Table B3). There was a negative trend in the incidences of adenoma or carcinoma (combined); however, the incidence in controls exceeded the previous historical control range (2% to 21%; Table B4d). The significance of this finding is uncertain.

Pituitary Gland: The incidences of pars distalis adenoma in 500 and 5,000 ppm males (20/50, 8/50, 14/50, 10/52) were significantly less than that in the controls; the trend was not significant (Table A3).

All Organs: At 2 years, the incidences of mononuclear cell leukemia in 1,700 and 5,000 ppm males were significantly less than that in the controls by the logistic regression test (37/50, 36/51, 20/51, 22/54; Table A3). However, the decreased incidences were not significant by the life table test (the most appropriate test for this generally fatal neoplasm) and were considered to be due primarily to reduced survival in these groups. Similar effects were not observed in female rats (Table B3).

GENETIC TOXICOLOGY

Results of mutagenicity tests with D&C Yellow No. 11 in *Salmonella typhimurium* were equivocal in one study, based on the responses observed in strain TA100 with 10% induced rat liver S9, and the results were weakly positive in a second study, which used slightly lower doses, based on responses observed in strains TA98 and TA100 with 30% induced rat or hamster liver S9 (Table C1; Zeiger *et al.*, 1988). No indication of mutagenic activity was observed in the absence of S9 in any of the strains tested. The data from the *S. typhimurium* studies indicate variable responses among replicate trials within a particular treatment condition; this may have been the result of precipitate formation at higher concentrations (333 µg/

plate and above) and consequent variability in the actual D&C Yellow No. 11 exposure concentrations.

In cytogenic tests with cultured Chinese hamster ovary cells, D&C Yellow No. 11 induced highly significant increases in both sister chromatid exchanges (Table C2) and chromosomal aberrations (Table C3) with and without S9. Cell cycle delay, requiring an extended incubation period, was observed in the sister chromatid exchange test at doses of 1.5 µg/mL and above; in the chromosomal aberrations test, no delay was observed in the absence of S9, but cultures treated in the presence of S9 were harvested late because cell cycle delay was anticipated. Less than 200 cells per dose level were scored in all but one dose level in the chromosomal aberrations test due to the high number of chromosomal aberrations per cell (cultures treated with S9), the frequency of aberrant cells, and the difficulty in finding scorable cells in some cases (Trial 1, without S9).

Despite the strong response seen in the *in vitro* chromosomal aberrations assay, no increase in the frequency of micronucleated normochromatic erythrocytes was observed in peripheral blood samples from male and female mice given D&C Yellow No. 11 in feed for 13 weeks (Table C4).

In conclusion, D&C Yellow No. 11 was mutagenic in bacteria and clastogenic in mammalian cells *in vitro*, but no evidence of clastogenicity was observed in the single *in vivo* study performed with male and female mice.

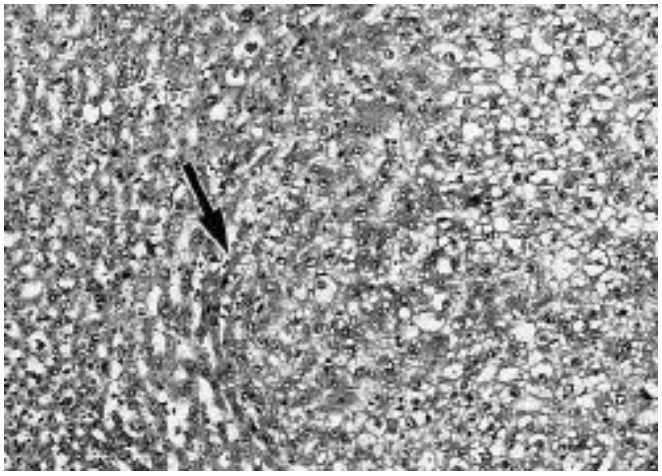


PLATE 1
Hepatocellular adenoma (arrow) in a female F344/N rat exposed to 500 ppm D&C Yellow No. 11 in feed for 2 years. Note the altered architecture and abruptly intersected and compressed normal hepatic cords (normal to left). H&E; 100×

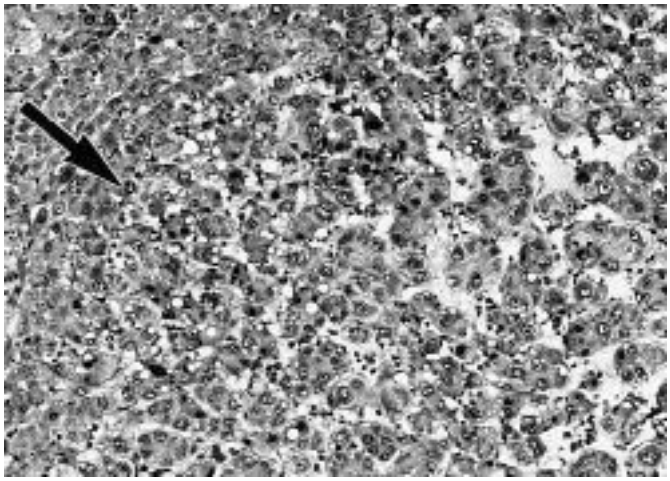


PLATE 2
Edge of a hepatocellular carcinoma (arrow) in a female F344/N rat exposed to 5,000 ppm D&C Yellow No. 11 in feed for 2 years. Note that neoplastic hepatocytes are in islands and clusters rather than cords. H&E; 100×

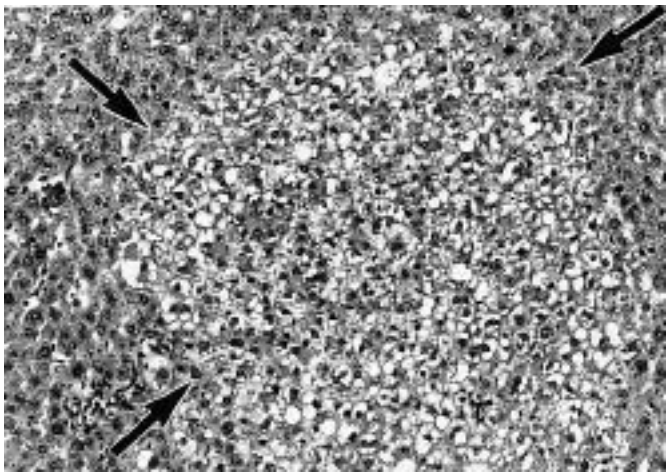


PLATE 3
Clear cell focus in the liver of a female F344/N rat exposed to 5,000 ppm D&C Yellow No. 11 in feed for 2 years. Note the hepatocytes within the roughly circular focus (arrows) are slightly larger and have relatively clear cytoplasm and central nuclei. H&E; 120×

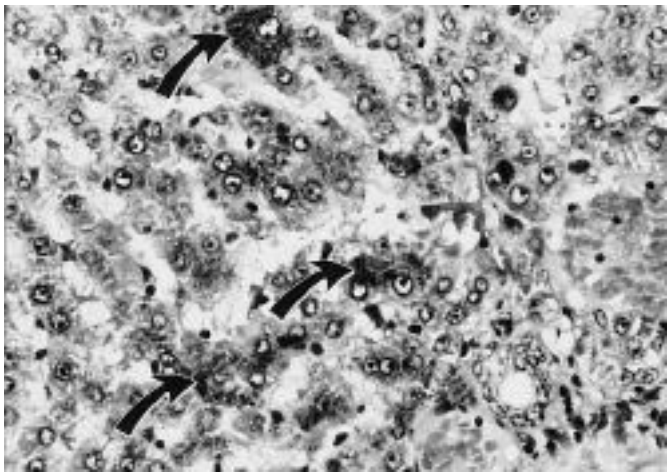


PLATE 4
Pigmentation (dark granular material) in hepatocytes (arrows) and Kupffer's cells in a male F344/N rat exposed to 5,000 ppm D&C Yellow No. 11 in feed for 2 years. H&E; 240×

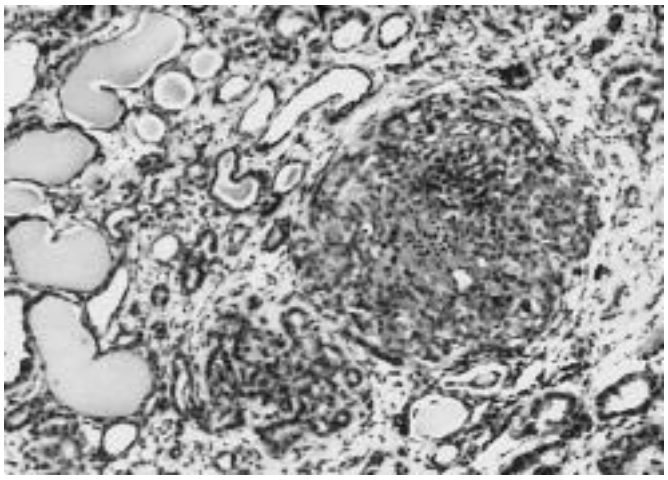


PLATE 5
Renal tubule adenoma in a male F344/N rat exposed to 1,700 ppm D&C Yellow No. 11 in feed for 2 years. Note the solid clusters of neoplastic epithelial cells in the connective tissue of a markedly nephrotic kidney. H&E; 100×

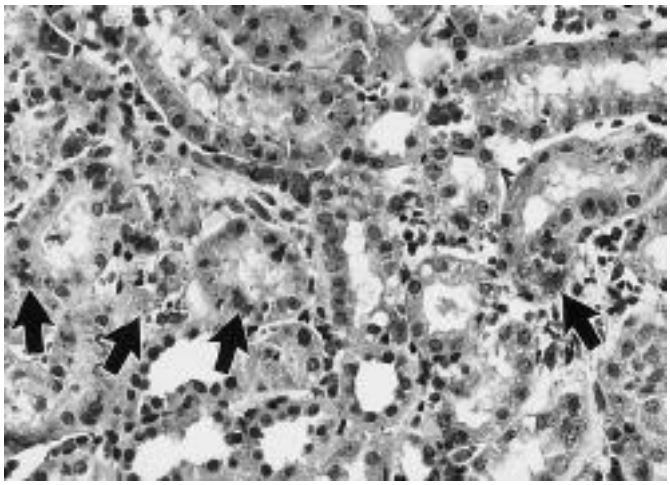


PLATE 6
Pigmentation in the kidney of a male F344/N rat exposed to 5,000 ppm D&C Yellow No. 11 in feed for 2 years. Note the granular dense material that stained brown in the cytoplasm of renal tubule epithelial cells (arrows). H&E; 240×

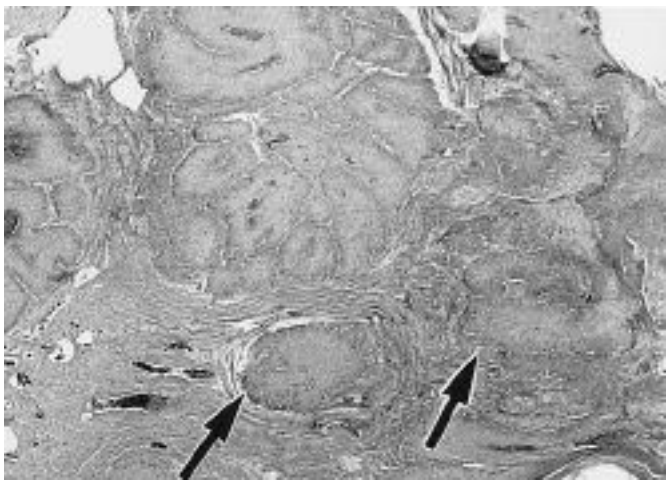


PLATE 7
Squamous cell carcinoma of the oral cavity in a male F344/N rat exposed to 5,000 ppm D&C Yellow No. 11 in feed for 2 years. Note that the nests of neoplastic squamous cells (arrows) invaded the underlying connective tissue. H&E; 30×

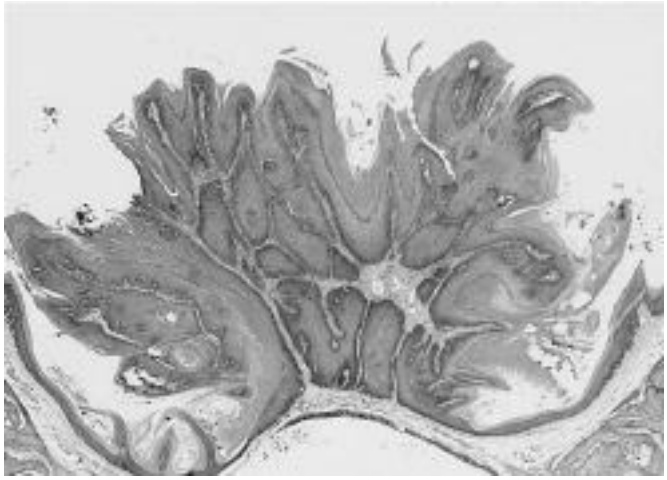


PLATE 8
Squamous cell papilloma of the oral mucosa in a female F344/N rat exposed to 500 ppm D&C Yellow No. 11 in feed for 2 years. Note that the super-ficially keratinized squamous epithelium proliferates on a branching fibrous connective tissue stalk. H&E; 17×

DISCUSSION AND CONCLUSIONS

D&C Yellow No. 11 was nominated for dosed-feed toxicity and carcinogenicity studies because it is a contaminant in D&C Yellow No. 10, a widely used, high-production dye which could potentially be ingested. Ingestion of D&C Yellow No. 11 has produced effects on growth in rats and mice. In the parental generation (F_0) in the current studies, mean body weight gains of exposed males and 5,000 ppm females were less than those of controls prior to cohabitation, but feed consumption by exposed rats was generally similar to that by the controls. There were no apparent D&C Yellow No. 11 effects on survival, reproductive performance (i.e., duration of gestation, average litter size, number of live pups per litter on days 4 or 21, or percentage of male pups), or mean litter weights at 1 or 4 days of age. However, by 14 days of age, the mean litter weights of all exposed groups were less than those of the controls. Although feed consumption by exposed and controls groups was similar, mean body weights of 1,700 and 5,000 ppm males and females were generally lower than those of controls throughout the 2-year study. A similar response was observed in the previous perinatal exposure study in F344/N rats (NTP, 1991a). In that study, the dams were given 0, 5,000, 17,000 or 50,000 ppm D&C Yellow No. 11 in feed for 4 weeks before mating (sires were not exposed) and throughout gestation, lactation, and weaning (day 28 after birthing). Pups were continued at the same exposure concentrations as their dams for 4 weeks after weaning. Mean pup body weights were similar at birth, but group mean body weights were less than those of the controls in all exposed groups at 4 weeks of age and in the 17,000 and 50,000 ppm groups at 8 weeks of age. However, in 13-week feed studies without perinatal exposure in F344/N rats and $B6C3F_1$ mice, up to 50,000 ppm D&C Yellow No. 11 in feed did not affect mean body weight gains or feed consumption (NTP, 1991a). The decreased body weight gains in rats are most likely a chemical-related phenomenon that affects optimum feed utilization (i.e., absorption and metabolism) rather than the result of decreased feed consumption. The effect of D&C Yellow No. 11 on body weight gains seems to be more pronounced when rats are exposed perinatally or in feed at a very young age.

After oral administration of radiolabeled D&C Yellow No. 11 to F344/N rats, the dye was rapidly absorbed and distributed to all tissues, and 98% was excreted within 72 hours (El Dareer *et al.*, 1988). D&C Yellow No. 11 was excreted in the feces and urine, and the liver and kidney had greater concentrations of radioactivity than did other tissues. More than 50% of an intravenous dose of D&C Yellow No. 11 was excreted via the bile 4 hours after administration (El Dareer *et al.*, 1988). No parent compound was recovered, and more than 10 metabolites were identified in the bile. The target organs in the current 2-year study are associated with known pathways of ingested D&C Yellow No. 11 [i.e., oral cavity (portal of entry), liver (metabolism), and bile duct and kidney (excretion)]. These organ sites were also the primary targets identified in short-term oral toxicity studies. Pigment was shown to accumulate in the liver, bile duct, and kidney of 8-week-old rats perinatally exposed followed by exposure to D&C Yellow No. 11 in feed after weaning and in dosed-feed studies in rats and mice with no perinatal exposure (NTP, 1991a; Eastin *et al.*, 1996). In addition, liver and kidney weights were increased in rats and mice after oral exposure to D&C Yellow No. 11, which suggests elevated metabolic and excretory activities in these organs.

Periportal degeneration of hepatocytes was apparent in rats examined at 8 weeks of age in the previous perinatal exposure study, in rats in the 14-day and 13-week studies (NTP, 1991a), and at the 12-month interim evaluation in the current study. Hepatocellular cytologic alteration (cytoplasmic basophilia and granularity) and pigmentation in less affected livers in the chronic studies also exhibited a periportal distribution. These alterations are clear indications of liver toxicity, but widespread hepatocellular necrosis was not seen at any time point studied. It was not possible to determine a zonal distribution of hepatocellular foci or neoplasms in the 2-year study. The

incidences of liver adenomas in 5,000 ppm males and females exceeded the historical control ranges.

The marginal indication of a neoplastic effect, coincident with exposure-related exacerbated severity of nephropathy and increased pigmentation of the renal tubule, in the standard evaluation (single sections) prompted an extended evaluation (step sections) of the kidney in male rats. The combined results of the standard and extended evaluations indicated a modest chemical-related increase in renal tubule neoplasms. The yellow-brown pigment was most likely D&C Yellow No. 11 or a metabolite because special stains of similar pigmentation in the 13-week studies were all negative for bile, hemosiderin, and lipofuscin. Cytoplasmic alteration (an increase in size and number of cytoplasmic hyaline droplets) was also present in the renal tubule epithelium in all exposed male groups. These hyaline droplets often formed large globules or irregularly shaped crystalline structures that stained similarly (Mallory-Heidenhain method) to the smaller granules of protein (α2μ-globulin) typically seen in the renal tubule cell cytoplasm of male F344/N rats. In a separate study (Eastin *et al.*, 1996), male rats given 5,000 ppm D&C Yellow No. 11 in feed for 70 days had cytoplasmic alteration and pigment in the renal tubules and hepatocellular degeneration and pigmentation similar to that seen at the same exposure concentration in the 13-week study. After a recovery period during which rats were given undosed feed for up to 28 days, pigment was still present in the liver biliary epithelium and renal tubule epithelium, and cytoplasmic alteration and pigment in the renal tubule epithelium were reduced in severity in all rats. There was no immunohistochemical evidence of an increase in the amount of α2μ-globulin in the kidney of rats with cytoplasmic alteration after 28 days on undosed feed; cytoplasmic hyaline droplets were similar in the controls. The cytoplasmic alteration, characterized by increased size and number of irregularly shaped hyaline droplets in the renal tubule epithelium of male F344/N rats, was morphologically similar to the abnormal accumulation of irregularly shaped hyaline droplets containing α2μ-globulin that has been described as a feature of chlorinated hydrocarbon ("hyaline droplet") nephropathy in male rats (NTP, 1991b; 1991c). Typically, chronic administration of chemicals causing this type of renal toxicity results in enhanced severity of nephropathy and an increase in proliferative lesions of the renal tubule epithelium in male rats. Other characteristic features of hydrocarbon nephropathy, including regeneration/necrosis, granular casts, and homogenous protein casts in renal tubules, were not observed in the current study. It appears that not all chemicals causing an increase in hyaline droplet accumulation may have the same spectrum of renal toxicity described for hydrocarbon nephropathy. Exposure to *p*-nitrobenzoic acid in feed for 13 weeks (NTP, 1994) also caused hyaline droplet accumulation in male rats; however, granular casts, necrosis, and regeneration were not evident, and there was no chemical-related exacerbation of nephropathy during the 2-year study.

D&C Yellow No. 11 has been shown to have skin sensitization and allergenic potential (Lamson *et al.*, 1982; Kita *et al.*, 1984). The finding of an association between D&C Yellow No. 11 exposure and oral cavity neoplasms in the current study was unexpected. However, it is possible that prolonged contact with a chemical shown to be a slight skin irritant and which has mutagenic and clastogenic activity with and without metabolic activation could have produced the response observed in the oral cavity. The numbers of papillomas of the tongue and oral mucosa in male rats are small, but the presence of an oral cavity carcinoma in one 500 ppm male at 12 months, in each of the exposed male groups at 2 years, plus carcinomas in two 5,000 ppm female rats at 2 years, as well as the low rates of these neoplasms in historical controls suggest that exposure to D&C Yellow No. 11 in feed is associated with neoplastic proliferation of the epithelium in the oral cavity. The incidence of squamous cell carcinoma in the oral cavity of 5,000 ppm females (2/51) exceeds the NTP historical control rate (4/1,301) and suggests that the neoplastic effect occurred in males and females.

The cause of the edema of the head and neck, which was observed grossly and resulted in the deaths of one male in the 1,700 ppm group and five males in the 5,000 ppm group, could not be determined. Possible causes of edema could include local obstruction of lymphatics or blood vasculature; heart failure; hypoproteinemia secondary to kidney or liver disease or intestinal malabsorption; or altered vascular pathways secondary to abnormal flow through the liver or to primary vasculopathy. Tissues from rats that had the diagnosis of edema were given complete histopathologic reviews without showing evidence of unusually

severe heart, liver, or kidney disease, or of vasculopathy or consistent intestinal mucosal hyperplasia. Since functional disturbances of fluid dynamics, heart function, or vascular tone do not necessarily have histopathologic correlates, the edema may have been a physiologic effect of D&C Yellow No. 11 or its metabolites.

CONCLUSIONS

Under the conditions of this perinatal exposure followed by a 2-year dosed feed study, there was *some evidence of carcinogenic activity** of D&C Yellow No. 11 in male F344/N rats based on increased incidences of hepatocellular adenoma, renal tubule neoplasms, and squamous cell neoplasms of the oral cavity. There was *some evidence of carcinogenic activity* in female F344/N rats based on increased incidences of hepatocellular neoplasms. Incidences of uncommon squamous cell carcinoma of the oral cavity in females may have been related to chemical treatment.

Exposure of rats to D&C Yellow No. 11 in feed for 2 years resulted in increased incidences of nonneoplastic liver lesions including clear cell foci, increased basophilia and granularity in the cytoplasm of hepatocytes, and bile duct, hepatocyte, and Kupffer cell pigmentation in males and females and mixed cell foci in males. In the kidney, there were increased incidences of renal tubule pigmentation and transitional epithelial hyperplasia in males and females and renal tubule hyperplasia in males. The severity of nephropathy was increased in exposed males and females.

---

* Explanation of Levels of Evidence of Carcinogenic Activity is on page 9. A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 11.

REFERENCES

Armitage, P. (1971). *Statistical Methods in Medical Research*, pp. 362-365. John Wiley and Sons, New York.

Ashby, J., and Tennant, R.W. (1991). Definitive relationships among chemical structure, carcinogenicity, and mutagenicity for 301 chemicals tested by the U.S. NTP. *Mutat. Res.* **257**, 229-306.

Björkner, B., and Magnusson, B. (1981). Patch test sensitization to D&C Yellow No. 11 and simultaneous reaction to Quinoline Yellow. *Contact Dermatitis* **7**, 1-4.

Björkner, B., and Niklasson, B. (1983). Contact allergic reaction to D&C Yellow No. 11 and Quinoline Yellow. *Contact Dermatitis* **9**, 263-268.

Boorman, G.A., Montgomery, C.A., Jr., Eustis, S.L., Wolfe, M.J., McConnell, E.E., and Hardisty, J.F. (1985). Quality assurance in pathology for rodent carcinogenicity studies. In *Handbook of Carcinogen Testing* (H.A. Milman and E.K. Weisburger, Eds.), pp. 345-357. Noyes Publications, Park Ridge, NJ.

Calnan, C.D. (1981). Quinazoline yellow dermatitis (D&C Yellow 11) in an eye cream. *Contact Dermatitis* **7**, 271.

Charbonneau, M., Lock, E.A., Strasser, J., Cox, M.G., Turner, M.J., and Bus, J.S. (1987). 2,2,4-Trimethylpentane-induced nephrotoxicity. I. Metabolic disposition of TMP in male and female Fischer 344 rats. *Toxicol. Appl. Pharmacol.* **91**, 171-181.

Code of Federal Regulations (CFR) **21**, Part 58.

Code of Federal Regulations (CFR) **21** §74.1711.

Colour Index (1982). Pigments and Solvent Dyes, 3rd ed. The Society of Dyers and Colourists, pp. 147-148.

The Cosmetic, Toiletry and Fragrance Association (CTFA) Cosmetic Ingredient Dictionary (1982). 3rd ed. (N.F. Estrin, P.A. Crosley, and C.R. Haynes, Eds.), p. 72. Washington, DC.

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

Crawford, B.D. (1985). Perspectives on the somatic mutation model of carcinogenesis. In *Advances in Modern Environmental Toxicology. Mechanisms and Toxicity of Chemical Carcinogens and Mutagens* (M.A. Mehlman, W.G. Flamm, and R.J. Lorentzen, Eds.), pp. 13-59. Princeton Scientific Publishing Co. Inc., Princeton, NJ.

Dinse, G.E., and Haseman, J.K. (1986). Logistic regression analysis of incidental-tumor data from animal carcinogenicity experiments. *Fundam. Appl. Toxicol.* **6**, 44-52.

Dinse, G.E., and Lagakos, S.W. (1983). Regression analysis of tumour prevalence data. *Appl. Statist.* **32**, 236-248.

Dixon, W.J., and Massey, F.J., Jr. (1951). *Introduction to Statistical Analysis*, 1st ed., pp. 145-147. McGraw-Hill Book Company, Inc., New York.

Dunn, O.J. (1964). Multiple comparisons using rank sums. *Technometrics* **6**, 241-252.

Dunnett, C.W. (1955). A multiple comparison procedure for comparing several treatments with a control. *J. Am. Stat. Assoc.* **50**, 1096-1121.

Eastin, W.C., Elwell, M.R., Grumbein, S., and Yuan, J.-H. (1996). Effect of D&C Yellow No. 11 ingestion on F344/N rats and B6C3F$_1$ mice. *J. Toxicol. Environ. Health* **48**, 101-117.

El Dareer, S.M., Kalin, J.R., Tillery, K.F., and Hill, D.L. (1988). Disposition of 2-(2-quinolyl)-1,3-indandione (D.C. Yellow # 11) in rats dosed orally or intravenously. *J. Toxicol. Environ. Health* **23**, 385-393.

Galloway, S.M., Armstrong, M.J., Reuben, C., Colman, S., Brown, B., Cannon, C., Bloom, A.D., Nakamura, F., Ahmed, M., Duk, S., Rimpo, J., Margolin, B.H., Resnick, M.A., Anderson, B., and Zeiger, E. (1987). Chromosome aberrations and sister chromatid exchanges in Chinese hamster ovary cells: Evaluations of 108 chemicals. *Environ. Mol. Mutagen.* **10** (Suppl. 10), 1-175.

Gart, J.J., Chu, K.C., and Tarone, R.E. (1979). Statistical issues in interpretation of chronic bioassay tests for carcinogenicity. *J. Natl. Cancer Inst.* **62**, 957-974.

Gershbein, L.L. (1982). Action of dyes and indicators on rat-liver regeneration. *Food Chem. Toxicol.* **20**, 1-8.

Hansen, W.H., Wilson, D.C., and Fitzhugh, O.G. (1960). Subacute oral toxicity of ten D&C coal-tar colors. *Fed. Proc.* **19**, 390.

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

Hollander, M., and Wolfe, D.A. (1973). *Nonparametric Statistical Methods*, pp. 120-123. John Wiley and Sons, New York.

Jonckheere, A.R. (1954). A distribution-free *k*-sample test against ordered alternatives. *Biometrika* **41**, 133-145.

Jordan, W.P., Jr. (1981). Contact dermatitis from D&C Yellow 11 dye in a toilet bar soap. *J. Am. Acad. Dermatol.* **4**, 613-614.

Kaplan, E.L., and Meier, P. (1958). Nonparametric estimation from incomplete observations. *J. Am. Stat. Assoc.* **53**, 457-481.

Kita, S., Kobayashi, T., Kutsuna, H., and Kligman, A.M. (1984). Human maximization testing of D&C Yellow No. 10 and Yellow No. 11. *Contact Dermatitis* **11**, 210-213.

Lamson, S.A., Kong, B.M., and De Salva, S.J. (1982). D&C Yellow Nos. 10 and 11: Delayed contact hypersensitivity in the guinea pig. *Contact Dermatitis* **8**, 200-203.

McConnell, E.E., Solleveld, H.A., Swenberg, J.A., and Boorman, G.A. (1986). Guidelines for combining neoplasms for evaluation of rodent carcinogenesis studies. *JNCI* **76**, 283-289.

MacGregor, J.T., Wehr, C.M., and Langlois, R.G. (1983). A simple fluorescent staining procedure for micronuclei and RNA in erythrocytes using Hoescht 33258 and pyronin Y. *Mutat. Res.* **120**, 269-275.

MacGregor, J.T., Wehr, C.M., Henika, P.R., and Shelby, M.D. (1990). The *in vivo* erythrocyte micronucleus test: Measurement at steady state increases assay efficiency and permits integration with toxicity studies. *Fundam. Appl. Toxicol.* **14**, 513-522.

McKnight, B., and Crowley, J. (1984). Tests for differences in tumor incidence based on animal carcinogenesis experiments. *J. Am. Stat. Assoc.* **79**, 639-648.

Marmion, D.M. (1991). *Handbook of U.S. Colorants: Foods, Drugs, Cosmetics, and Medical Devices*, 3rd ed., pp. 26-31. John Wiley and Sons, New York.

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

Matsunaga, K., Hosokawa, K., Suzuki, M., Arima, Y., and Hayakawa, R. (1988). Occupational allergic contact dermatitis in beauticians. *Contact Dermatitis* **18**, 94-96.

The Merck Index (1989). 11th ed. (S. Budavari, Ed.), p. 1286. Merck and Company, Rahway, NJ.

Meyer, M., Brock, K., Lawrence, K., Casto, B., and Moore, M.M. (1986). Evaluation of the effect of agar on the results obtained in the L5178Y mouse lymphoma assay. *Environ. Mutagen.* **8**, 727-740.

Miller, J.A., and Miller, E.C. (1977). Ultimate chemical carcinogens as reactive mutagenic electrophiles. In *Origins of Human Cancer* (H.H. Hiatt, J.D. Watson, and J.A. Winsten, Eds.), pp. 605-627. Cold Spring Harbor Laboratory, Cold Spring Harbor, NY.

Moore, M.M., Allen, J.W., Claxton, L., Doerr, C., Gwaltney, C., Dutcher, J.S., Kohan, M., Lawrence, B.K., Templeton, R., and Westbrook-Collins, B. (1988). Mutagenic screening of marker grenade dyes by the *Salmonella* reversion assay, L5178Y/TK$^{+/-}$ mouse lymphoma assay, and in vivo sister chromatid exchange analysis in mice. *Environ. Mol. Mutagen.* **12**, 219-233.

National Cancer Institute (NCI) (1976). Guidelines for Carcinogen Bioassay in Small Rodents. Technical Report Series No. 1. NIH Publication No. 76-801. U.S. Department of Health, Education, and Welfare, Public Health Service, National Institutes of Health, Bethesda, MD.

National Institute for Occupational Safety and Health (NIOSH) (1990). National Occupational Exposure Survey (1981-1983), unpublished provisional data as of July 1, 1990. NIOSH, Cincinnati, OH.

National Institutes of Health (NIH) (1978). Open Formula Rat and Mouse Ration (NIH-07). Specification NIH-11-1335. U.S. Department of Health, Education, and Welfare, Public Health Service, National Institutes of Health, Bethesda, MD.

National Toxicology Program (NTP) (1991a). Toxicity Studies of D&C Yellow No. 11 in F344/N Rats and B6C3F$_1$ Mice (Feed Studies). Toxicity Report Series No. 8. NIH Publication No. 91-3127. U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health, Research Triangle Park, NC.

National Toxicology Program (NTP) (1991b). Toxicity Studies of 1,2,4,5-Tetrachlorobenzene in F344/N Rats and B6C3F$_1$ Mice (Feed Studies). Toxicity Report Series No. 7. NIH Publication No. 91-3126. U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health, Research Triangle Park, NC.

National Toxicology Program (NTP) (1991c). Toxicity Studies of Pentachlorobenzene (CAS No. 608-93-5) in F344/N Rats and B6C3F$_1$ Mice (Feed Studies). Toxicity Report Series No. 6. NIH Publication No. 91-3125. U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health, Research Triangle Park, NC.

National Toxicology Program (NTP) (1994). Toxicology and Carcinogenesis Studies of *p*-Nitrobenzoic Acid (CAS No. 62-23-7) in F344/N Rats and B6C3F$_1$ Mice (Feed Studies). Technical Report Series No. 442. NIH Publication No. 95-3358. U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health, Research Triangle Park, NC.

Palazzolo, M.J., and DiPasquale, L.C. (1983). The sensitization potential of D&C Yellow No. 11 in guinea pigs. *Contact Dermatitis* **9**, 367-371.

Rapaport, M.J. (1984). Allergy to yellow dyes. *Arch. Dermatol.* **120**, 535-536.

Sato, Y., Kutsuna, H., Kobayashi, T., and Mitsui, T. (1984). D&C Nos. 10 and 11: Chemical composition analysis and delayed contact hypersensitivity testing in the guinea pig. *Contact Dermatitis* **10**, 30-38.

Schmid, W. (1976). The micronucleus test for cytogenetic analysis. In *Chemical Mutagens: Principles and Methods for their Detection* (A. Hollaender, Ed.), Vol. 4, pp. 31-53. Plenum Press, New York.

Shirley, E. (1977). A non-parametric equivalent of Williams' test for contrasting increasing dose levels of a treatment. *Biometrics* **33**, 386-389.

Straus, D.S. (1981). Somatic mutation, cellular differentiation, and cancer causation. *JNCI* **67**, 233-241.

Sun, J.D., Henderson, R.F., Marshall, T.C., Cheng, Y.-S., Dutcher, J.S., Pickrell, J.A., Mauderly, J.L., Hahn, F.F., Banas, D.A., Seiler, F.A., and Hobbs, C.H. (1987). The inhalation toxicity of two commercial dyes: Solvent Yellow 33 and Solvent Green 3. *Fundam. Appl. Toxicol.* **8**, 358-371.

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

Tennant, R.W., Margolin, B.H., Shelby, M.D., Zeiger, E., Haseman, J.K., Spalding, J., Caspary, W., Resnick, M., Stasiewicz, S., Anderson, B., and Minor, R. (1987). Prediction of chemical carcinogenicity in rodents from *in vitro* genetic toxicity assays. *Science* **236**, 933-941.

Weaver, J.E. (1983). Dose response relationships in delayed hypersensitivity to quinoline dyes. *Contact Dermatitis* **9**, 309-312.

Williams, D.A. (1971). A test for differences between treatment means when several dose levels are compared with a zero dose control. *Biometrics* **27**, 103-117.

Williams, D.A. (1972). The comparison of several dose levels with a zero dose control. *Biometrics* **28**, 519-531.

Yuan, J., Jameson, C.W., Goehl, T.J., Elwell, M.R., Leininger, J.R., Thompson, M.B., Corniffe, G., and Carlton, T. (1992). Application of molecular encapsulation for toxicology studies: Comparative toxicity of *p*-chloro-α,α,α-trifluorotoluene in α-cyclodextrin vehicle versus corn oil vehicle in male and female Fischer 344 rats and B6C3F1 mice. *Fundam. Appl. Toxicol.* **18**, 460-470.

Zeiger, E., Anderson, B., Haworth, S., Lawlor, T., and Mortelmans, K. (1988). *Salmonella* mutagenicity tests: IV. Results from the testing of 300 chemicals. *Environ. Mol. Mutagen.* **11** (Suppl. 12), 1-158.

Zeiger, E., Haseman, J.K., Shelby, M.D., Margolin, B.H., and Tennant, R.W. (1990). Evaluation of four in vitro genetic toxicity tests for predicting rodent carcinogenicity: Confirmation of earlier results with 41 additional chemicals. *Environ. Mol. Mutagen.* **16** (Suppl. 18), 1-14.

APPENDIX A
SUMMARY OF LESIONS IN MALE RATS IN THE 2-YEAR FEED STUDY OF D&C YELLOW NO. 11

TABLE A1	Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11	55
TABLE A2	Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11	60
TABLE A3	Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11	80
TABLE A4a	Historical Incidence of Hepatocellular Neoplasms in Untreated Male F344/N Rats	88
TABLE A4b	Historical Incidence of Renal Tubule Neoplasms in Untreated Male F344/N Rats	88
TABLE A4c	Historical Incidence of Oral Cavity Neoplasms in Untreated Male F344/N Rats	89
TABLE A4d	Historical Incidence of Testicular Adenoma in Untreated Male F344/N Rats	89
TABLE A5	Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11	90

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Disposition Summary				
Animals initially in study	60	60	60	60
12-Month interim evaluation	10	9	9	6
Early deaths				
Moribund	29	29	41	49
Natural deaths	2	2	1	3
Other			1	
Survivors				
Terminal sacrifice	19	20	8	2
Animals examined microscopically	60	60	60	60
12-Month Interim Evaluation				
Alimentary System				
Pancreas	(10)	(9)	(9)	(6)
Acinar cell, adenoma				1 (17%)
Tongue		(1)		
Squamous cell carcinoma		1 (100%)		
Endocrine System				
Pituitary gland	(10)	(9)	(8)	(6)
Pars distalis, adenoma	1 (10%)			
Genital System				
Testes	(10)	(9)	(9)	(6)
Interstitial cell, adenoma		3 (33%)	1 (11%)	
Systems Examined With No Neoplasms Observed				
Cardiovascular System				
General Body System				
Hematopoietic System				
Integumentary System				
Musculoskeletal System				
Nervous System				
Respiratory System				
Special Senses System				
Urinary System				
2-Year Study				
Alimentary System				
Intestine large, colon	(50)	(50)	(50)	(54)
Polyp adenomatous			1 (2%)	
Intestine large, cecum	(50)	(50)	(50)	(54)
Hemangioma				1 (2%)
Intestine small, jejunum	(50)	(50)	(51)	(54)
Carcinoma	1 (2%)			1 (2%)

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Alimentary System (continued)				
Intestine small, ileum	(49)	(50)	(50)	(54)
Carcinoma				1 (2%)
Liver	(50)	(51)	(51)	(54)
Hepatocellular adenoma	1 (2%)	2 (4%)	1 (2%)	6 (11%)
Hepatocellular adenoma, multiple				1 (2%)
Mesentery	(11)	(12)	(9)	(13)
Lipoma	1 (9%)			
Oral mucosa		(2)	(3)	(3)
Squamous cell carcinoma		1 (50%)	1 (33%)	1 (33%)
Squamous cell papilloma		1 (50%)	1 (33%)	2 (67%)
Pancreas	(50)	(51)	(51)	(54)
Acinar cell, adenoma		1 (2%)	4 (8%)	1 (2%)
Salivary glands	(50)	(51)	(50)	(54)
Carcinoma, metastatic, Zymbal's gland			1 (2%)	
Stomach, forestomach	(50)	(51)	(51)	(54)
Squamous cell papilloma		2 (4%)		
Stomach, glandular	(50)	(51)	(51)	(54)
Carcinoma		1 (2%)		
Tongue	(1)		(1)	(4)
Squamous cell papilloma	1 (100%)		1 (100%)	2 (50%)
Cardiovascular System				
Heart	(50)	(51)	(51)	(54)
Schwannoma malignant	1 (2%)			
Endocrine System				
Adrenal cortex	(50)	(51)	(51)	(54)
Adrenal medulla	(50)	(50)	(51)	(54)
Neuroblastoma malignant			1 (2%)	
Pheochromocytoma malignant	4 (8%)	1 (2%)	2 (4%)	
Pheochromocytoma benign	7 (14%)	8 (16%)	11 (22%)	6 (11%)
Bilateral, pheochromocytoma benign			1 (2%)	2 (4%)
Islets, pancreatic	(50)	(51)	(51)	(54)
Adenoma	4 (8%)	3 (6%)		
Carcinoma	1 (2%)		2 (4%)	
Pituitary gland	(50)	(50)	(50)	(52)
Pars distalis, adenoma	20 (40%)	7 (14%)	13 (26%)	8 (15%)
Pars distalis, adenoma, multiple		1 (2%)	1 (2%)	2 (4%)
Pars intermedia, adenoma		1 (2%)	1 (2%)	1 (2%)
Thyroid gland	(50)	(51)	(50)	(54)
C-cell, adenoma	5 (10%)	2 (4%)	3 (6%)	3 (6%)
C-cell, adenoma, multiple	1 (2%)	1 (2%)		
C-cell, carcinoma			1 (2%)	1 (2%)
Follicular cell, adenoma			1 (2%)	
Follicular cell, carcinoma	3 (6%)	1 (2%)	1 (2%)	3 (6%)
General Body System				
Peritoneum	(2)	(1)	(3)	(3)

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Genital System				
Epididymis	(49)	(51)	(51)	(54)
Preputial gland	(49)	(50)	(51)	(54)
Adenoma	5 (10%)	2 (4%)	1 (2%)	
Carcinoma	5 (10%)	4 (8%)	1 (2%)	2 (4%)
Prostate	(50)	(51)	(51)	(53)
Adenocarcinoma		1 (2%)		
Seminal vesicle	(50)	(51)	(51)	(54)
Testes	(49)	(51)	(51)	(54)
Bilateral, interstitial cell, adenoma	30 (61%)	36 (71%)	42 (82%)	39 (72%)
Interstitial cell, adenoma	9 (18%)	10 (20%)	6 (12%)	7 (13%)
Hematopoietic System				
Bone marrow	(50)	(51)	(51)	(54)
Lymph node	(20)	(23)	(26)	(45)
Renal, pheochromocytoma malignant, metastatic, adrenal medulla	1 (5%)			
Lymph node, mandibular	(50)	(51)	(50)	(53)
Carcinoma, metastatic, Zymbal's gland			1 (2%)	
Lymph node, mesenteric	(50)	(50)	(51)	(54)
Spleen	(50)	(51)	(51)	(54)
Hemangiosarcoma			1 (2%)	
Thymus	(48)	(49)	(49)	(53)
Integumentary System				
Mammary gland	(48)	(48)	(49)	(53)
Fibroadenoma	3 (6%)	2 (4%)	3 (6%)	1 (2%)
Skin	(50)	(50)	(51)	(54)
Basal cell adenoma	1 (2%)	1 (2%)	2 (4%)	
Basal cell carcinoma			2 (4%)	
Keratoacanthoma	7 (14%)	4 (8%)	2 (4%)	1 (2%)
Squamous cell papilloma	3 (6%)	3 (6%)	2 (4%)	3 (6%)
Trichoepithelioma	1 (2%)			
Sebaceous gland, adenoma	1 (2%)			
Sebaceous gland, carcinoma	1 (2%)			1 (2%)
Subcutaneous tissue, fibroma	3 (6%)	2 (4%)	3 (6%)	
Subcutaneous tissue, lipoma		1 (2%)		
Subcutaneous tissue, schwannoma malignant	1 (2%)			
Musculoskeletal System				
Bone	(50)	(51)	(51)	(54)
Osteosarcoma	1 (2%)			
Skeletal muscle	(1)	(2)	(5)	(9)
Nervous System				
Brain	(50)	(51)	(51)	(54)
Hemangioma	1 (2%)			
Spinal cord	(1)	(2)	(1)	

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Respiratory System				
Lung	(50)	(51)	(51)	(54)
Alveolar/bronchiolar adenoma	7 (14%)	1 (2%)	2 (4%)	1 (2%)
Alveolar/bronchiolar adenoma, multiple		1 (2%)		
Alveolar/bronchiolar carcinoma	1 (2%)	1 (2%)		1 (2%)
Carcinoma, metastatic, Zymbal's gland			1 (2%)	
Osteosarcoma, metastatic, bone	1 (2%)			
Special Senses System				
Zymbal's gland			(1)	(1)
Carcinoma			1 (100%)	1 (100%)
Urinary System				
Kidney	(50)	(51)	(51)	(54)
Osteosarcoma, metastatic, bone	1 (2%)			
Renal tubule, adenoma				2 (4%)
Renal tubule, carcinoma			1 (2%)	
Urinary bladder	(50)	(51)	(51)	(54)
Systemic Lesions				
Multiple organs[b]	(50)	(51)	(51)	(54)
Leukemia mononuclear	37 (74%)	36 (71%)	20 (39%)	22 (41%)
Lymphoma malignant	1 (2%)			
Mesothelioma malignant	2 (4%)	2 (4%)	3 (6%)	4 (7%)

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Neoplasm Summary				
Total animals with primary neoplasms[c]				
12-Month interim evaluation	1	4	1	1
2-Year study	50	51	50	49
Total primary neoplasms				
12-Month interim evaluation	1	4	1	1
2-Year study	170	140	139	127
Total animals with benign neoplasms				
12-Month interim evaluation	1	3	1	1
2-Year study	49	50	49	49
Total benign neoplasms				
12-Month interim evaluation	1	3	1	1
2-Year study	111	92	102	89
Total animals with malignant neoplasms				
12-Month interim evaluation		1		
2-Year study	42	39	30	26
Total malignant neoplasms				
12-Month interim evaluation		1		
2-Year study	59	48	37	38
Total animals with metastatic neoplasms				
2-Year study	2		1	
Total metastatic neoplasms				
2-Year study	3		3	

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm

Number of Days on Study	481	509	530	590	600	615	619	621	622	623	628	632	632	637	638	650	656	656	667	677	688	693	695	695	701
Carcass ID Number	053	011	001	054	044	058	028	039	045	040	029	004	034	015	047	018	032	050	005	016	036	017	007	037	008
Alimentary System																									
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Intestine small, ileum	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hepatocellular adenoma																									
Mesentery	+								+		+					+					+				
Lipoma																									
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tongue																									
Squamous cell papilloma																									
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Schwannoma malignant																								X	
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma malignant																				X					
Pheochromocytoma benign						X							X							X					
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																									
Carcinoma																									
Parathyroid gland	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma			X		X			X		X	X			X		X		X	X					X	
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-cell, adenoma				X																			X		
C-cell, adenoma, multiple					X																				
Follicular cell, carcinoma																							X		
General Body System																									
Peritoneum																	+								+
Genital System																									
Epididymis	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Preputial gland	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma															X										X
Carcinoma																					X				

+: Tissue examined microscopically
A: Autolysis precludes examination
M: Missing tissue
I: Insufficient tissue
X: Lesion present
Blank: Not examined

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	702	706	706	716	719	722	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	734	734		
Carcass ID Number	043	003	024	031	020	052	006	009	010	012	014	019	022	023	025	027	030	033	035	041	048	055	056	059	060	Total Tissues/ Tumors
Alimentary System																										
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma																							X			1
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Hepatocellular adenoma																		X								1
Mesentery	+									+				+							+			+	+	11
Lipoma																									X	1
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Tongue																								+		1
Squamous cell papilloma																								X		1
Cardiovascular System																										
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Schwannoma malignant																										1
Endocrine System																										
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pheochromocytoma malignant													X								X				X	4
Pheochromocytoma benign				X					X									X				X				7
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adenoma				X													X					X			X	4
Carcinoma													X													1
Parathyroid gland	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	47
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pars distalis, adenoma							X	X	X		X		X	X			X		X				X	X		20
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
C-cell, adenoma	X																		X		X					5
C-cell, adenoma, multiple																										1
Follicular cell, carcinoma									X													X				3
General Body System																										
Peritoneum																										2
Genital System																										
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Adenoma															X			X							X	5
Carcinoma							X	X				X					X									5

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	481	509	530	590	600	615	619	621	622	623	628	632	632	637	638	650	656	656	667	677	688	693	695	695	701
Carcass ID Number	053	011	001	054	044	058	028	039	045	040	029	004	034	015	047	018	032	050	005	016	036	017	007	037	008
Genital System (continued)																									
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Testes	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Bilateral, interstitial cell, adenoma				X					X			X	X		X		X			X	X	X	X	X	X
Interstitial cell, adenoma	X	X				X										X									
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node		+	+		+			+	+				+		+	+	+			+	+	+	+		
Renal, pheochromocytoma malignant, metastatic, adrenal medulla																				X					
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Thymus	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	M	+	+	+	+	+
Fibroadenoma																									
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Basal cell adenoma																								X	
Keratoacanthoma											X		X				X				X		X		
Squamous cell papilloma																									
Trichoepithelioma																									
Sebaceous gland, adenoma																									
Sebaceous gland, carcinoma																									X
Subcutaneous tissue, fibroma												X	X												
Subcutaneous tissue, schwannoma malignant						X																			
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma				X																					
Skeletal muscle																				+					
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hemangioma																									
Peripheral nerve								+																	
Spinal cord								+																	
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma																X	X								
Alveolar/bronchiolar carcinoma																									
Osteosarcoma, metastatic, bone				X																					
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	702	706	706	716	719	722	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	734	734		
Carcass ID Number	043	003	024	031	020	052	006	009	010	012	014	019	022	023	025	027	030	033	035	041	048	055	056	059	060	Total Tissues/ Tumors
Genital System (continued)																										
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Bilateral, interstitial cell, adenoma	X		X	X	X	X	X	X	X	X	X	X			X	X				X	X		X	X	X	30
Interstitial cell, adenoma		X												X			X		X			X				9
Hematopoietic System																										
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node	+	+		+	+				+								+	+								20
Renal, pheochromocytoma malignant, metastatic, adrenal medulla																										1
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	48
Integumentary System																										
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Fibroadenoma				X						X		X														3
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Basal cell adenoma																										1
Keratoacanthoma																	X						X			7
Squamous cell papilloma			X			X												X								3
Trichoepithelioma								X																		1
Sebaceous gland, adenoma																				X						1
Sebaceous gland, carcinoma																										1
Subcutaneous tissue, fibroma																		X								3
Subcutaneous tissue, schwannoma malignant																									1	
Musculoskeletal System																										
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Osteosarcoma																										1
Skeletal muscle																										1
Nervous System																										
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Hemangioma																						X				1
Peripheral nerve																										1
Spinal cord																										1
Respiratory System																										
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Alveolar/bronchiolar adenoma				X					X	X	X									X						7
Alveolar/bronchiolar carcinoma																									X	1
Osteosarcoma, metastatic, bone																										1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	481	509	530	590	600	615	619	621	622	623	628	632	632	637	638	650	656	656	667	677	688	693	695	695	701
Carcass ID Number	053	011	001	054	044	058	028	039	045	040	029	004	034	015	047	018	032	050	005	016	036	017	007	037	008
Special Senses System																									
Ear																									
Eye																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma, metastatic, bone				X																					
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear	X	X	X		X	X	X	X	X	X		X	X	X	X	X			X	X	X	X	X	X	X
Lymphoma malignant													X												
Mesothelioma malignant																	X								X

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	702	706	706	716	719	722	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	734	734	
Carcass ID Number	043	003	024	031	020	052	006	009	010	012	014	019	022	023	025	027	030	033	035	041	048	055	056	059	060	Total Tissues/ Tumors
Special Senses System																										
Ear	+												+													2
Eye													+													1
Urinary System																										
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Osteosarcoma, metastatic, bone																										1
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Systemic Lesions																										
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Leukemia mononuclear	X	X	X	X	X	X		X	X		X	X					X		X	X	X		X		X	37
Lymphoma malignant																										1
Mesothelioma malignant																										2

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm

Number of Days on Study	229	509	509	510	545	546	558	558	560	570	575	586	593	602	611	628	628	642	656	656	658	672	677	688	695
Carcass ID Number	101	070	075	117	104	073	064	092	097	099	091	106	074	116	062	094	108	066	103	120	061	086	088	087	090
Alimentary System																									
Esophagus	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hepatocellular adenoma																			X						
Mesentery			+													+				+		+			
Oral mucosa																					+				
Squamous cell carcinoma																									
Squamous cell papilloma																					X				
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Acinar cell, adenoma																									
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Squamous cell papilloma																									
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Tooth																									
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma malignant																									
Pheochromocytoma benign									X								X				X	X			X
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma												X		X											
Parathyroid gland	+	M	+	+	+	+	M	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pituitary gland	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma												X		X				X				X			
Pars distalis, adenoma, multiple																									
Pars intermedia, adenoma																									
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-cell, adenoma											X														
C-cell, adenoma, multiple																									
Follicular cell, carcinoma							X																		
General Body System																									
Peritoneum																									
Genital System																									
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+
Adenoma																									
Carcinoma				X																			X	X	

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm (continued)

Number of Days on Study	712	716	719	719	726	727	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	734	734	
Carcass ID Number	110	084	080	098	096	078	065	067	068	072	077	079	081	082	089	093	095	100	105	107	109	112	113	114	115	119	Total Tissues/ Tumors
Alimentary System																											
Esophagus	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	I	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	I	I	+	+	+	+	+	+	+	+	+	+	+	49
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Hepatocellular adenoma																		X									2
Mesentery		+		+		+			+		+				+				+						+		12
Oral mucosa	+																										2
Squamous cell carcinoma	X																										1
Squamous cell papilloma																											1
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Acinar cell, adenoma																							X				1
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Squamous cell papilloma	X													X													2
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Carcinoma																			X								1
Tooth																						+					1
Cardiovascular System																											
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Endocrine System																											
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	50
Pheochromocytoma malignant																			X								1
Pheochromocytoma benign	X					X														X							8
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenoma		X																									3
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	47
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pars distalis, adenoma					X		X																			X	7
Pars distalis, adenoma, multiple											X																1
Pars intermedia, adenoma																					X						1
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
C-cell, adenoma																	X										2
C-cell, adenoma, multiple																								X			1
Follicular cell, carcinoma																											1
General Body System																											
Peritoneum						+																					1
Genital System																											
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adenoma					X									X													2
Carcinoma																										X	4

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm (continued)

Number of Days on Study	22	09	09	10	45	46	58	58	60	70	75	86	93	02	11	28	28	42	56	56	58	72	77	88	95
Carcass ID Number	101	070	075	117	104	073	064	092	097	099	091	106	074	116	062	094	108	066	103	120	061	086	088	087	090
Genital System (continued)																									
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenocarcinoma		X																							
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Bilateral, interstitial cell, adenoma		X		X		X				X	X		X		X	X	X			X	X		X	X	X
Interstitial cell, adenoma			X		X			X	X										X			X			
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node	+	+			+	+	+		+				+		+	+				+		+	+		+
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M
Lymph node, mediastinal	+																								
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Thymus	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	M	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
Fibroadenoma																									
Skin	+	+	+	+	+	+	+	I	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Basal cell adenoma																						X			
Keratoacanthoma																					X				
Squamous cell papilloma																									
Subcutaneous tissue, fibroma							X																		
Subcutaneous tissue, lipoma																									
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Skeletal muscle																		+							
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Peripheral nerve										+															
Spinal cord										+															
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma																									
Alveolar/bronchiolar adenoma, multiple																									
Alveolar/bronchiolar carcinoma							X																		
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Special Senses System																									
Ear																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear	X	X	X		X	X	X	X	X	X	X	X	X		X	X			X	X	X	X	X	X	X
Mesothelioma malignant																									

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm (continued)

Number of Days on Study	712	716	719	719	726	727	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	733	734	734	
Carcass ID Number	1010	1084	0080	0098	0096	0078	0065	0067	0068	0072	0077	0079	0081	0082	0089	0093	0095	0100	0105	0107	0109	0112	0113	0114	0115	0119	Total Tissues/ Tumors
Genital System (continued)																											
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenocarcinoma																											1
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Bilateral, interstitial cell, adenoma	X	X		X		X		X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	36
Interstitial cell, adenoma			X		X		X										X										10
Hematopoietic System																											
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node	+	+		+		+					+		+		+				+				+			+	23
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node, mediastinal																											1
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Integumentary System																											
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Fibroadenoma									X										X								2
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Basal cell adenoma																											1
Keratoacanthoma							X		X														X				4
Squamous cell papilloma											X										X				X		3
Subcutaneous tissue, fibroma	X																										2
Subcutaneous tissue, lipoma																		X									1
Musculoskeletal System																											
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Skeletal muscle															+												2
Nervous System																											
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Peripheral nerve			+																								2
Spinal cord			+																								2
Respiratory System																											
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Alveolar/bronchiolar adenoma																						X					1
Alveolar/bronchiolar adenoma, multiple																		X									1
Alveolar/bronchiolar carcinoma																											1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Special Senses System																											
Ear				+																							1
Urinary System																											
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Systemic Lesions																											
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Leukemia mononuclear	X	X		X	X	X								X	X		X	X	X	X	X	X	X		X		36
Mesothelioma malignant						X					X																2

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm

Number of Days on Study	246	384	406	453	468	491	499	502	551	590	600	600	607	610	615	631	646	646	649	649	649	650	650	659	663
Carcass ID Number	175	179	170	139	157	126	180	178	145	173	146	163	166	136	162	168	147	165	128	134	174	161	171	138	131
Alimentary System																									
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	I	+	M	+	+	+	+	+	+	+	+
Intestine large, colon	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Polyp adenomatous																									
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hepatocellular adenoma													X												
Mesentery												+			+		+				+				+
Oral mucosa			+																						
Squamous cell carcinoma																									
Squamous cell papilloma			X																						
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Acinar cell, adenoma																				X					
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
Carcinoma, metastatic, Zymbal's gland		X																							
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tongue																									
Squamous cell papilloma																									
Tooth																									
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Neuroblastoma malignant										X															
Pheochromocytoma malignant																									
Pheochromocytoma benign							X	X	X										X					X	
Bilateral, pheochromocytoma benign																					X				
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Parathyroid gland	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	M	+	+	+	+	M	+	+	+
Pituitary gland	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma								X			X							X	X	X	X		X		
Pars distalis, adenoma, multiple																									
Pars intermedia, adenoma																									
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
C-cell, adenoma														X											
C-cell, carcinoma																									
Follicular cell, adenoma																								X	
Follicular cell, carcinoma																									
General Body System																									
Peritoneum						+					+														

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	663	667	672	680	686	686	686	686	692	695	695	695	699	706	712	714	719	727	733	733	733	733	733	733	733	733	
Carcass ID Number	151	155	176	148	133	142	153	154	167	144	159	177	127	169	140	130	152	123	121	125	129	137	149	150	160	164	Total Tissues/ Tumors
Alimentary System																											
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Polyp adenomatous																		X									1
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine large, cecum	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, ileum	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Hepatocellular adenoma																											1
Mesentery		+				+															+			+			9
Oral mucosa				+														+									3
Squamous cell carcinoma				X																							1
Squamous cell papilloma																											1
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Acinar cell, adenoma			X							X		X															4
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma, metastatic, Zymbal's gland																											1
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Tongue						+																					1
Squamous cell papilloma						X																					1
Tooth				+				+						+													3
Cardiovascular System																											
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Endocrine System																											
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Neuroblastoma malignant																											1
Pheochromocytoma malignant															X								X				2
Pheochromocytoma benign								X	X				X			X	X									X	11
Bilateral, pheochromocytoma benign																											1
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Carcinoma							X												X								2
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pars distalis, adenoma		X	X			X														X				X	X		13
Pars distalis, adenoma, multiple												X															1
Pars intermedia, adenoma																	X										1
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
C-cell, adenoma							X	X																			3
C-cell, carcinoma																				X							1
Follicular cell, adenoma																											1
Follicular cell, carcinoma																					X						1
General Body System																											
Peritoneum				+																							3

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	246	384	406	453	468	491	499	502	551	590	600	600	607	610	615	631	646	646	649	649	649	650	650	659	663
Carcass ID Number	175	179	170	139	157	126	180	178	145	173	146	163	166	136	162	168	147	165	128	134	174	161	171	138	131
Genital System																									
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																									
Carcinoma																							X		
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Bilateral, interstitial cell, adenoma						X	X	X	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	X
Interstitial cell, adenoma				X	X													X							
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node			+	+	+	+		+	+	+	+					+		+	+		+			+	+
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
Carcinoma, metastatic, Zymbal's gland		X																							
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hemangiosarcoma																									
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+
Fibroadenoma																									
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Basal cell adenoma										X															
Basal cell carcinoma																	X								
Keratoacanthoma																							X		
Squamous cell papilloma																									
Subcutaneous tissue, fibroma																									
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Skeletal muscle								+	+		+					+									
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Peripheral nerve																									
Spinal cord																									
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma				X																					
Carcinoma, metastatic, Zymbal's gland		X																							
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+
Special Senses System																									
Ear																									
Zymbal's gland		+																							
Carcinoma		X																							

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	663	667	672	680	686	686	686	686	692	695	695	695	699	706	712	714	719	727	733	733	733	733	733	733	733	733	
Carcass ID Number	151	155	176	148	133	142	153	154	167	144	159	177	127	169	140	130	152	123	121	125	129	137	149	150	160	164	Total Tissues/ Tumors
Genital System																											
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenoma						X																					1
Carcinoma																											1
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Bilateral, interstitial cell, adenoma		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X		X	X	X		X	X	42
Interstitial cell, adenoma	X																			X				X			6
Hematopoietic System																											
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node	+			+			+	+	+	+	+					+	+	+	+						+		26
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma, metastatic, Zymbal's gland																											1
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Hemangiosarcoma																						X					1
Thymus	+	+	+	+	M	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Integumentary System																											
Mammary gland	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Fibroadenoma									X										X		X						3
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Basal cell adenoma											X																2
Basal cell carcinoma																				X							2
Keratoacanthoma															X												2
Squamous cell papilloma						X											X										2
Subcutaneous tissue, fibroma	X															X									X		3
Musculoskeletal System																											
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Skeletal muscle																+											5
Nervous System																											
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Peripheral nerve									+																		1
Spinal cord									+																		1
Respiratory System																											
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Alveolar/bronchiolar adenoma														X													2
Carcinoma, metastatic, Zymbal's gland																											1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Special Senses System																											
Ear			+					M																			1
Zymbal's gland																											1
Carcinoma																											1

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	246	384	406	453	468	491	499	502	551	590	600	600	607	610	615	631	646	646	649	649	649	650	650	659	663
Carcass ID Number	175	179	170	139	157	126	180	178	145	173	146	163	166	136	162	168	147	165	128	134	174	161	171	138	131
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Renal tubule, carcinoma																									
Urethra																									
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear				X	X					X		X	X		X	X	X	X			X			X	X
Mesothelioma malignant						X					X														

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	663	667	672	680	686	686	686	686	692	695	695	695	699	706	712	714	719	727	733	733	733	733	733	733	733	733	
Carcass ID Number	151	155	176	148	133	142	153	154	167	144	159	177	227	169	140	130	152	223	121	125	129	137	149	150	160	164	Total Tissues/ Tumors
Urinary System																											
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Renal tubule, carcinoma					X																						1
Urethra	+																										1
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Systemic Lesions																											
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Leukemia mononuclear	X			X		X			X	X					X					X				X			20
Mesothelioma malignant				X																							3

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm

Number of Days on Study	302	320	345	345	370	432	481	492	496	498	498	498	502	505	509	510	534	546	563	566	568	572	589	590	596
Carcass ID Number	230	197	198	227	240	224	219	187	201	186	195	203	226	221	228	238	182	192	233	193	196	184	229	234	185
Alimentary System																									
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hemangioma																									
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hepatocellular adenoma												X							X						
Hepatocellular adenoma, multiple																									
Mesentery				+	+						+		+												+
Oral mucosa							+																	+	
Squamous cell carcinoma																								X	
Squamous cell papilloma							X																		
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Acinar cell, adenoma																									
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tongue																									
Squamous cell papilloma																									
Cardiovascular System																									
Blood vessel	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma benign												X													
Bilateral, pheochromocytoma benign																									
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	M	+	+	+	+	+	+	+
Pituitary gland	+	+	M	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma															X							X			
Pars distalis, adenoma, multiple																				X			X		
Pars intermedia, adenoma																									
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-cell, adenoma											X														
C-cell, carcinoma																									
Follicular cell, carcinoma																									X
General Body System																									
Peritoneum									+																
Genital System																									
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	596	602	615	617	624	624	629	656	656	658	678	678	690	691	693	695	695	695	700	700	704	706	716	716	722	726	727	733	733	
Carcass ID Number	188	217	210	204	189	190	237	199	216	212	232	235	239	215	208	194	220	236	207	209	214	223	205	206	211	218	222	220	225	Total Tissues/ Tumors
Alimentary System																														
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Hemangioma					X																									1
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Carcinoma																											X			1
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Carcinoma																			X											1
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Hepatocellular adenoma															X								X			X			X	6
Hepatocellular adenoma, multiple																						X								1
Mesentery		+										+	+		+				+	+		+				+				13
Oral mucosa																+														3
Squamous cell carcinoma																														1
Squamous cell papilloma																X														2
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Acinar cell, adenoma												X																		1
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Tongue						+											+						+	+						4
Squamous cell papilloma																	X							X						2
Cardiovascular System																														
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	53
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Endocrine System																														
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Pheochromocytoma benign						X			X						X	X				X										6
Bilateral, pheochromocytoma benign													X														X			2
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	52
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	52
Pars distalis, adenoma		X		X	X		X									X				X										8
Pars distalis, adenoma, multiple																														2
Pars intermedia, adenoma																		X												1
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
C-cell, adenoma																				X				X						3
C-cell, carcinoma		X																												1
Follicular cell, carcinoma	X																										X			3
General Body System																														
Peritoneum										+														+						3
Genital System																														
Epididymis	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Preputial gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Carcinoma																								X		X				2

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	302	320	345	345	370	432	481	492	496	498	498	498	502	505	509	510	534	546	563	566	568	572	589	590	596
Carcass ID Number	230	197	198	227	240	224	219	187	201	186	195	203	226	221	228	238	182	192	233	193	196	184	229	234	185
Genital System (continued)																									
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Bilateral, interstitial cell, adenoma								X	X	X	X	X	X	X			X	X	X	X	X			X	X
Interstitial cell, adenoma					X		X									X						X			
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node		+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mandibular	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mediastinal		+		+																					
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Fibroadenoma																									
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Keratoacanthoma																									X
Squamous cell papilloma																									
Sebaceous gland, carcinoma																									
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Skeletal muscle			+	+		+				+	+	+				+		+					+		
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma																									
Alveolar/bronchiolar carcinoma																									
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Special Senses System																									
Ear																									
Eye		+																							
Zymbal's gland																									
Carcinoma																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Renal tubule, adenoma																									
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear							X					X	X							X	X		X		X
Mesothelioma malignant									X												X				

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	596	602	615	617	624	624	629	656	656	658	678	678	690	691	693	695	695	695	700	700	704	706	716	716	722	726	727	733	733	
Carcass ID Number	188	117	210	204	189	190	237	199	216	212	232	235	239	215	208	194	220	236	207	209	214	223	205	206	211	218	222	220	225	Total Tissues/ Tumors
Genital System (continued)																														
Prostate	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+		+	+	+	+	53
Seminal vesicle	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Testes	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Bilateral, interstitial cell, adenoma	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	39
Interstitial cell, adenoma					X		X							X																7
Hematopoietic System																														
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Lymph node		+	+	+		+		+	+	+	+	+	+	+	+	+		+	+	+		+		+	+		+	+	+	45
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	53
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Lymph node, mediastinal																														2
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Thymus	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	53
Integumentary System																														
Mammary gland	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	53
Fibroadenoma																									X					1
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Keratoacanthoma																														1
Squamous cell papilloma						X																X							X	3
Sebaceous gland, carcinoma															X															1
Musculoskeletal System																														
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Skeletal muscle																														9
Nervous System																														
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Respiratory System																														
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Alveolar/bronchiolar adenoma																	X													1
Alveolar/bronchiolar carcinoma																								X						1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Special Senses System																														
Ear																	+													1
Eye																														1
Zymbal's gland								+																						1
Carcinoma								X																						1
Urinary System																														
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Renal tubule, adenoma										X															X					2
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Systemic Lesions																														
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	54
Leukemia mononuclear	X		X					X	X	X			X	X	X		X		X		X	X		X	X	X				22
Mesothelioma malignant										X														X						4

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Adrenal Medulla: Benign Pheochromocytoma				
Overall rate[a]	7/50 (14%)	8/50 (16%)	12/51 (24%)	8/54 (15%)
Adjusted rate[b]	25.9%	25.8%	49.3%	55.7%
Terminal rate[c]	3/19 (16%)	1/19 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	615	560	499	498
Life table test[d]	P=0.025	P=0.483	P=0.028	P=0.032
Logistic regression test[d]	P=0.377	P=0.469	P=0.143	P=0.258
Cochran-Armitage test[d]	P=0.533N			
Fisher exact test[d]		P=0.500	P=0.166	P=0.565
Adrenal Medulla: Malignant Pheochromocytoma				
Overall rate	4/50 (8%)	1/50 (2%)	2/51 (4%)	0/54 (0%)
Adjusted rate	18.5%	5.3%	19.8%	0.0%
Terminal rate	3/19 (16%)	1/19 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	677	733 (T)	712	—[e]
Life table test	P=0.454N	P=0.182N	P=0.663	P=0.404N
Logistic regression test	P=0.255N	P=0.186N	P=0.574N	P=0.211N
Cochran-Armitage test	P=0.072N			
Fisher exact test		P=0.181N	P=0.329N	P=0.050N
Adrenal Medulla: Benign or Malignant Pheochromocytoma				
Overall rate	10/50 (20%)	9/50 (18%)	14/51 (27%)	8/54 (15%)
Adjusted rate	39.8%	29.9%	60.2%	55.7%
Terminal rate	6/19 (32%)	2/19 (11%)	2/8 (25%)	0/2 (0%)
First incidence (days)	615	560	499	498
Life table test	P=0.033	P=0.516N	P=0.024	P=0.051
Logistic regression test	P=0.521	P=0.558N	P=0.167	P=0.456
Cochran-Armitage test	P=0.286N			
Fisher exact test		P=0.500N	P=0.260	P=0.330N
Kidney (Renal Tubule): Adenoma (Step Sections)				
Overall rate	0/50 (0%)	2/51 (4%)	4/51 (8%)	2/54 (4%)
Adjusted rate	0.0%	7.1%	22.5%	18.8%
Terminal rate	0/19 (0%)	1/20 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	—	558	649	678
Life table test	P=0.080	P=0.241	P=0.027	P=0.090
Logistic regression test	P=0.259	P=0.255	P=0.046	P=0.120
Cochran-Armitage test	P=0.406			
Fisher exact test		P=0.252	P=0.061	P=0.267
Kidney (Renal Tubule): Adenoma (Single and Step Sections)				
Overall rate	0/50 (0%)	2/51 (4%)	4/51 (8%)	4/54 (7%)
Adjusted rate	0.0%	7.1%	22.5%	38.3%
Terminal rate	0/19 (0%)	1/20 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	—	558	649	658
Life table test	P=0.003	P=0.241	P=0.027	P=0.006
Logistic regression test	P=0.032	P=0.255	P=0.046	P=0.014
Cochran-Armitage test	P=0.106			
Fisher exact test		P=0.252	P=0.061	P=0.069

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Kidney (Renal Tubule): Adenoma or Carcinoma (Single and Step Sections)				
Overall rate	0/50 (0%)	2/51 (4%)	5/51 (10%)	4/54 (7%)
Adjusted rate	0.0%	7.1%	26.1%	38.3%
Terminal rate	0/19 (0%)	1/20 (5%)	1/8 (13%)	0/2 (0%)
First incidence (days)	—	558	649	658
Life table test	P=0.004	P=0.241	P=0.012	P=0.006
Logistic regression test	P=0.036	P=0.255	P=0.022	P=0.014
Cochran-Armitage test	P=0.121			
Fisher exact test		P=0.252	P=0.030	P=0.069
Liver: Hepatocellular Adenoma				
Overall rate	1/50 (2%)	2/51 (4%)	1/51 (2%)	7/54 (13%)
Adjusted rate	5.3%	7.9%	2.6%	75.0%
Terminal rate	1/19 (5%)	1/20 (5%)	0/8 (0%)	1/2 (50%)
First incidence (days)	733 (T)	656	607	498
Life table test	P<0.001	P=0.508	P=0.650	P<0.001
Logistic regression test	P=0.001	P=0.487	P=0.757	P=0.008
Cochran-Armitage test	P=0.007			
Fisher exact test		P=0.508	P=0.748N	P=0.038
Lung: Alveolar/bronchiolar Adenoma				
Overall rate	7/50 (14%)	2/51 (4%)	2/51 (4%)	1/54 (2%)
Adjusted rate	28.9%	10.0%	9.6%	7.1%
Terminal rate	4/19 (21%)	2/20 (10%)	0/8 (0%)	0/2 (0%)
First incidence (days)	650	733 (T)	453	695
Life table test	P=0.405N	P=0.073N	P=0.294N	P=0.414N
Logistic regression test	P=0.112N	P=0.084N	P=0.103N	P=0.148N
Cochran-Armitage test	P=0.045N			
Fisher exact test		P=0.075N	P=0.075N	P=0.023N
Lung: Alveolar/bronchiolar Adenoma or Carcinoma				
Overall rate	8/50 (16%)	3/51 (6%)	2/51 (4%)	2/54 (4%)
Adjusted rate	33.7%	12.0%	9.6%	20.4%
Terminal rate	5/19 (26%)	2/20 (10%)	0/8 (0%)	0/2 (0%)
First incidence (days)	650	558	453	695
Life table test	P=0.574N	P=0.092N	P=0.233N	P=0.642N
Logistic regression test	P=0.155N	P=0.111N	P=0.069N	P=0.260N
Cochran-Armitage test	P=0.063N			
Fisher exact test		P=0.094N	P=0.043N	P=0.035N
Mammary Gland: Fibroadenoma				
Overall rate	3/50 (6%)	2/51 (4%)	3/51 (6%)	1/54 (2%)
Adjusted rate	14.6%	10.0%	29.2%	20.0%
Terminal rate	2/19 (11%)	2/20 (10%)	2/8 (25%)	0/2 (0%)
First incidence (days)	716	733 (T)	692	722
Life table test	P=0.285	P=0.469N	P=0.285	P=0.573
Logistic regression test	P=0.532	P=0.470N	P=0.394	P=0.729
Cochran-Armitage test	P=0.247N			
Fisher exact test		P=0.491N	P=0.652N	P=0.280N

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Oral Cavity (Oral Mucosa or Tongue): Squamous Cell Papilloma				
Overall rate	1/50 (2%)	1/51 (2%)	2/51 (4%)	4/54 (7%)
Adjusted rate	5.3%	3.2%	6.5%	28.1%
Terminal rate	1/19 (5%)	0/20 (0%)	0/8 (0%)	0/2 (0%)
First incidence (days)	733 (T)	658	406	481
Life table test	P=0.008	P=0.758N	P=0.367	P=0.020
Logistic regression test	P=0.087	P=0.755	P=0.606	P=0.110
Cochran-Armitage test	P=0.088			
Fisher exact test		P=0.748N	P=0.508	P=0.206
Oral Cavity (Oral Mucosa or Tongue): Squamous Cell Papilloma or Squamous Cell Carcinoma				
Overall rate	1/50 (2%)	2/51 (4%)	3/51 (6%)	5/54 (9%)
Adjusted rate	5.3%	6.9%	10.6%	30.4%
Terminal rate	1/19 (5%)	0/20 (0%)	0/8 (0%)	0/2 (0%)
First incidence (days)	733 (T)	658	406	481
Life table test	P=0.004	P=0.521	P=0.191	P=0.008
Logistic regression test	P=0.066	P=0.487	P=0.369	P=0.069
Cochran-Armitage test	P=0.078			
Fisher exact test		P=0.508	P=0.316	P=0.121
Pancreas: Adenoma				
Overall rate	0/50 (0%)	1/51 (2%)	4/51 (8%)	1/54 (2%)
Adjusted rate	0.0%	5.0%	18.0%	5.3%
Terminal rate	0/19 (0%)	1/20 (5%)	0/8 (0%)	0/2 (0%)
First incidence (days)	—	733 (T)	649	678
Life table test	P=0.236	P=0.510	P=0.031	P=0.409
Logistic regression test	P=0.368	P=0.510	P=0.048	P=0.432
Cochran-Armitage test	P=0.540			
Fisher exact test		P=0.505	P=0.061	P=0.519
Pancreatic Islets: Adenoma				
Overall rate	4/50 (8%)	3/51 (6%)	0/51 (0%)	0/54 (0%)
Adjusted rate	19.6%	8.9%	0.0%	0.0%
Terminal rate	3/19 (16%)	0/20 (0%)	0/8 (0%)	0/2 (0%)
First incidence (days)	716	586	—	—
Life table test	P=0.146N	P=0.494N	P=0.215N	P=0.483N
Logistic regression test	P=0.050N	P=0.514N	P=0.167N	P=0.320N
Cochran-Armitage test	P=0.028N			
Fisher exact test		P=0.489N	P=0.056N	P=0.050N
Pancreatic Islets: Adenoma or Carcinoma				
Overall rate	5/50 (10%)	3/51 (6%)	2/51 (4%)	0/54 (0%)
Adjusted rate	24.6%	8.9%	16.5%	0.0%
Terminal rate	4/19 (21%)	0/20 (0%)	1/8 (13%)	0/2 (0%)
First incidence (days)	716	586	686	—
Life table test	P=0.222N	P=0.350N	P=0.597N	P=0.430N
Logistic regression test	P=0.066N	P=0.374N	P=0.479N	P=0.265N
Cochran-Armitage test	P=0.024N			
Fisher exact test		P=0.346N	P=0.219N	P=0.023N

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Pituitary Gland (Pars Distalis): Adenoma				
Overall rate	20/50 (40%)	8/50 (16%)	14/50 (28%)	10/52 (19%)
Adjusted rate	63.9%	27.7%	58.0%	36.1%
Terminal rate	10/19 (53%)	3/20 (15%)	3/8 (38%)	0/2 (0%)
First incidence (days)	530	586	502	509
Life table test	P=0.129	P=0.014N	P=0.492	P=0.342
Logistic regression test	P=0.160N	P=0.008N	P=0.214N	P=0.046N
Cochran-Armitage test	P=0.098N			
Fisher exact test		P=0.007N	P=0.146N	P=0.018N
Preputial Gland: Adenoma				
Overall rate	5/49 (10%)	2/50 (4%)	1/51 (2%)	0/54 (0%)
Adjusted rate	21.3%	9.3%	4.5%	0.0%
Terminal rate	3/19 (16%)	1/20 (5%)	0/8 (0%)	0/2 (0%)
First incidence (days)	638	726	686	—
Life table test	P=0.200N	P=0.203N	P=0.287N	P=0.302N
Logistic regression test	P=0.083N	P=0.229N	P=0.167N	P=0.107N
Cochran-Armitage test	P=0.026N			
Fisher exact test		P=0.210N	P=0.093N	P=0.022N
Preputial Gland: Carcinoma				
Overall rate	5/49 (10%)	4/50 (8%)	1/51 (2%)	2/54 (4%)
Adjusted rate	23.7%	13.4%	3.3%	35.7%
Terminal rate	4/19 (21%)	1/20 (5%)	0/8 (0%)	0/2 (0%)
First incidence (days)	688	510	650	716
Life table test	P=0.548	P=0.496N	P=0.318N	P=0.355
Logistic regression test	P=0.327N	P=0.522N	P=0.192N	P=0.645
Cochran-Armitage test	P=0.142N			
Fisher exact test		P=0.487N	P=0.093N	P=0.180N
Preputial Gland: Adenoma or Carcinoma				
Overall rate	10/49 (20%)	6/50 (12%)	2/51 (4%)	2/54 (4%)
Adjusted rate	42.9%	21.7%	7.7%	35.7%
Terminal rate	7/19 (37%)	2/20 (10%)	0/8 (0%)	0/2 (0%)
First incidence (days)	638	510	650	716
Life table test	P=0.365N	P=0.199N	P=0.138N	P=0.638N
Logistic regression test	P=0.068N	P=0.233N	P=0.044N	P=0.184N
Cochran-Armitage test	P=0.010N			
Fisher exact test		P=0.194N	P=0.011N	P=0.009N
Skin: Squamous Cell Papilloma				
Overall rate	3/50 (6%)	3/51 (6%)	2/51 (4%)	3/54 (6%)
Adjusted rate	13.7%	15.0%	14.1%	58.0%
Terminal rate	1/19 (5%)	3/20 (15%)	0/8 (0%)	1/2 (50%)
First incidence (days)	706	733 (T)	686	624
Life table test	P=0.036	P=0.633N	P=0.577	P=0.101
Logistic regression test	P=0.236	P=0.652N	P=0.674N	P=0.359
Cochran-Armitage test	P=0.571N			
Fisher exact test		P=0.652N	P=0.491N	P=0.623N

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Skin: Keratoacanthoma				
Overall rate	7/50 (14%)	4/51 (8%)	2/51 (4%)	1/54 (2%)
Adjusted rate	23.1%	17.7%	11.4%	3.3%
Terminal rate	2/19 (11%)	3/20 (15%)	0/8 (0%)	0/2 (0%)
First incidence (days)	628	658	650	596
Life table test	P=0.244N	P=0.276N	P=0.232N	P=0.236N
Logistic regression test	P=0.064N	P=0.279N	P=0.105N	P=0.054N
Cochran-Armitage test	P=0.025N			
Fisher exact test		P=0.251N	P=0.075N	P=0.023N
Skin: Squamous Cell Papilloma or Keratoacanthoma				
Overall rate	10/50 (20%)	7/51 (14%)	4/51 (8%)	4/54 (7%)
Adjusted rate	34.1%	32.3%	23.9%	59.4%
Terminal rate	3/19 (16%)	6/20 (30%)	0/8 (0%)	1/2 (50%)
First incidence (days)	628	658	650	596
Life table test	P=0.369	P=0.293N	P=0.335N	P=0.544
Logistic regression test	P=0.282N	P=0.327N	P=0.134N	P=0.230N
Cochran-Armitage test	P=0.059N			
Fisher exact test		P=0.282N	P=0.069N	P=0.055N
Skin: Trichoepithelioma, Basal Cell Adenoma, or Basal Cell Carcinoma				
Overall rate	2/50 (4%)	1/51 (2%)	4/51 (8%)	0/54 (0%)
Adjusted rate	8.6%	3.3%	21.9%	0.0%
Terminal rate	1/19 (5%)	0/20 (0%)	1/8 (13%)	0/2 (0%)
First incidence (days)	695	672	590	—
Life table test	P=0.545N	P=0.502N	P=0.156	P=0.551N
Logistic regression test	P=0.322N	P=0.513N	P=0.280	P=0.416N
Cochran-Armitage test	P=0.216N			
Fisher exact test		P=0.492N	P=0.348	P=0.229N
Skin: Squamous Cell Papilloma, Keratoacanthoma, Trichoepithelioma, Basal Cell Adenoma, or Basal Cell Carcinoma				
Overall rate	12/50 (24%)	8/51 (16%)	8/51 (16%)	4/54 (7%)
Adjusted rate	40.5%	34.5%	40.6%	59.4%
Terminal rate	4/19 (21%)	6/20 (30%)	1/8 (13%)	1/2 (50%)
First incidence (days)	628	658	590	596
Life table test	P=0.441	P=0.229N	P=0.516	P=0.595N
Logistic regression test	P=0.171N	P=0.255N	P=0.359N	P=0.135N
Cochran-Armitage test	P=0.025N			
Fisher exact test		P=0.213N	P=0.213N	P=0.018N
Skin (Subcutaneous Tissue): Fibroma				
Overall rate	3/50 (6%)	2/51 (4%)	3/51 (6%)	0/54 (0%)
Adjusted rate	10.1%	6.0%	23.4%	0.0%
Terminal rate	1/19 (5%)	0/20 (0%)	1/8 (13%)	0/2 (0%)
First incidence (days)	632	558	663	—
Life table test	P=0.359N	P=0.509N	P=0.442	P=0.333N
Logistic regression test	P=0.140N	P=0.468N	P=0.602	P=0.141N
Cochran-Armitage test	P=0.097N			
Fisher exact test		P=0.491N	P=0.652N	P=0.108N

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Testes: Adenoma				
Overall rate	39/49 (80%)	46/51 (90%)	48/51 (94%)	46/54 (85%)
Adjusted rate	95.0%	100.0%	100.0%	100.0%
Terminal rate	17/19 (89%)	20/20 (100%)	8/8 (100%)	2/2 (100%)
First incidence (days)	481	509	453	370
Life table test	P<0.001	P=0.212	P<0.001	P<0.001
Logistic regression test	P=0.018	P=0.033	P<0.001	P=0.005
Cochran-Armitage test	P=0.533			
Fisher exact test		P=0.114	P=0.030	P=0.313
Thyroid Gland (C-cell): Adenoma				
Overall rate	6/50 (12%)	3/51 (6%)	3/50 (6%)	3/54 (6%)
Adjusted rate	20.7%	12.2%	11.5%	23.8%
Terminal rate	2/19 (11%)	2/20 (10%)	0/8 (0%)	0/2 (0%)
First incidence (days)	590	575	610	498
Life table test	P=0.373	P=0.265N	P=0.484N	P=0.525
Logistic regression test	P=0.379N	P=0.242N	P=0.274N	P=0.325N
Cochran-Armitage test	P=0.267N			
Fisher exact test		P=0.234N	P=0.243N	P=0.207N
Thyroid Gland (C-cell): Adenoma or Carcinoma				
Overall rate	6/50 (12%)	3/51 (6%)	4/50 (8%)	4/54 (7%)
Adjusted rate	20.7%	12.2%	22.5%	26.5%
Terminal rate	2/19 (11%)	2/20 (10%)	1/8 (13%)	0/2 (0%)
First incidence (days)	590	575	610	498
Life table test	P=0.157	P=0.265N	P=0.594	P=0.350
Logistic regression test	P=0.568N	P=0.242N	P=0.436N	P=0.445N
Cochran-Armitage test	P=0.419N			
Fisher exact test		P=0.234N	P=0.370N	P=0.322N
Thyroid Gland (Follicular Cell): Carcinoma				
Overall rate	3/50 (6%)	1/51 (2%)	1/50 (2%)	3/54 (6%)
Adjusted rate	13.7%	2.2%	12.5%	37.8%
Terminal rate	2/19 (11%)	0/20 (0%)	1/8 (13%)	0/2 (0%)
First incidence (days)	695	558	733 (T)	596
Life table test	P=0.055	P=0.305N	P=0.602N	P=0.144
Logistic regression test	P=0.321	P=0.307N	P=0.514N	P=0.499
Cochran-Armitage test	P=0.449			
Fisher exact test		P=0.301N	P=0.309N	P=0.623N
Thyroid Gland (Follicular Cell): Adenoma or Carcinoma				
Overall rate	3/50 (6%)	1/51 (2%)	2/50 (4%)	3/54 (6%)
Adjusted rate	13.7%	2.2%	15.6%	37.8%
Terminal rate	2/19 (11%)	0/20 (0%)	1/8 (13%)	0/2 (0%)
First incidence (days)	695	558	659	596
Life table test	P=0.063	P=0.305N	P=0.590	P=0.144
Logistic regression test	P=0.333	P=0.307N	P=0.662N	P=0.499
Cochran-Armitage test	P=0.463			
Fisher exact test		P=0.301N	P=0.500N	P=0.623N

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
All Organs: Mononuclear Cell Leukemia				
Overall rate	37/50 (74%)	36/51 (71%)	20/51 (39%)	22/54 (41%)
Adjusted rate	80.0%	77.2%	61.7%	81.6%
Terminal rate	10/19 (53%)	10/20 (50%)	2/8 (25%)	0/2 (0%)
First incidence (days)	481	222	453	481
Life table test	P=0.191	P=0.524N	P=0.241N	P=0.151
Logistic regression test	P<0.001N	P=0.358N	P<0.001N	P=0.011N
Cochran-Armitage test	P<0.001N			
Fisher exact test		P=0.436N	P<0.001N	P<0.001N
All Organs: Malignant Mesothelioma				
Overall rate	2/50 (4%)	2/51 (4%)	3/51 (6%)	4/54 (7%)
Adjusted rate	6.7%	9.5%	8.7%	22.7%
Terminal rate	0/19 (0%)	1/20 (5%)	0/8 (0%)	0/2 (0%)
First incidence (days)	656	727	491	496
Life table test	P=0.037	P=0.681N	P=0.384	P=0.127
Logistic regression test	P=0.258	P=0.682	P=0.592	P=0.363
Cochran-Armitage test	P=0.263			
Fisher exact test		P=0.684N	P=0.509	P=0.377
All Organs: Benign Neoplasms				
Overall rate	49/50 (98%)	50/51 (98%)	49/51 (96%)	49/54 (91%)
Adjusted rate	100.0%	100.0%	100.0%	100.0%
Terminal rate	19/19 (100%)	20/20 (100%)	8/8 (100%)	2/2 (100%)
First incidence (days)	481	509	406	370
Life table test	P<0.001	P=0.492	P=0.008	P<0.001
Logistic regression test	P=0.575	P=0.488	P=0.482	P=0.427
Cochran-Armitage test	P=0.034N			
Fisher exact test		P=0.748	P=0.508N	P=0.121N
All Organs: Malignant Neoplasms				
Overall rate	42/50 (84%)	39/51 (76%)	30/51 (59%)	26/54 (48%)
Adjusted rate	87.3%	82.1%	89.6%	88.8%
Terminal rate	13/19 (68%)	12/20 (60%)	6/8 (75%)	0/2 (0%)
First incidence (days)	481	222	384	481
Life table test	P=0.065	P=0.424N	P=0.417	P=0.080
Logistic regression test	P<0.001N	P=0.189N	P=0.005N	P=0.005N
Cochran-Armitage test	P<0.001N			
Fisher exact test		P=0.243N	P=0.005N	P<0.001N

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
All Organs: Benign or Malignant Neoplasms				
Overall rate	50/50 (100%)	51/51 (100%)	50/51 (98%)	49/54 (91%)
Adjusted rate	100.0%	100.0%	100.0%	100.0%
Terminal rate	19/19 (100%)	20/20 (100%)	8/8 (100%)	2/2 (100%)
First incidence (days)	481	222	384	370
Life table test	P<0.001	P=0.487	P=0.009	P<0.001
Logistic regression test	P=0.018N	—[f]	—	P=0.973N
Cochran-Armitage test	P=0.002N			
Fisher exact test		P=1.000N	P=0.505N	P=0.034N

(T)Terminal sacrifice
[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal gland, kidney, liver, lung, pancreas, pancreatic islets, pituitary gland, preputial gland, testes, and thyroid gland; for other tissues, denominator is number of animals necropsied.
[b] Kaplan-Meier estimated neoplasm incidence at the end of the study after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] Beneath the control incidence are the P values associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the controls and that exposed group. The life table test regards neoplasms in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The logistic regression test regards these lesions as nonfatal. The Cochran-Armitage and Fisher exact tests compare directly the overall incidence rates. For all tests, a negative trend or a lower incidence in an exposure group is indicated by **N**.
[e] Not applicable; no neoplasms in animal group
[f] Value of statistic cannot be computed.

TABLE A4a
Historical Incidence of Hepatocellular Neoplasms in Untreated Male F344/N Rats [a]

Study	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	0/51	0/51	0/51
Benzyl Acetate	5/50	1/50	5/50
Butyl Benzyl Phthalate	2/50	0/50	2/50
C.I. Pigment Red 23	2/50	1/50	3/50
C.I. Pigment Red 3	0/50	0/50	0/50
o-Nitroanisole	0/50	0/50	0/50
p-Nitrobenzoic Acid	2/49	3/49	4/49
Polysorbate 80	2/50	0/50	2/50
Overall Historical Incidence			
Total	30/1,301 (2.3%)	9/1,301 (0.7%)	37/1,301 (2.8%)
Standard deviation	2.9%	1.4%	3.3%
Range	0%-10%	0%-6%	0%-10%

[a] Data as of 12 May 1995

TABLE A4b
Historical Incidence of Renal Tubule Neoplasms in Untreated Male F344/N Rats [a]

Study	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	0/51	0/51	0/51
Benzyl Acetate	0/50	0/50	0/50
Butyl Benzyl Phthalate	1/50	0/50	1/50
C.I. Pigment Red 23	0/50	0/50	0/50
C.I. Pigment Red 3	0/50	1/50	1/50
o-Nitroanisole	0/49	0/49	0/49
p-Nitrobenzoic Acid	0/50	0/50	0/50
Polysorbate 80	0/50	1/50	1/50
Overall Historical Incidence			
Total	9/1,301 (0.7%)	3/1,301 (0.2%)	12/1,301 (0.9%)
Standard deviation	1.5%	0.7%	1.5%
Range	0%-6%	0%-2%	0%-6%

[a] Data as of 12 May 1995

TABLE A4c
Historical Incidence of Oral Cavity Neoplasms in Untreated Male F344/N Rats [a]

Study	Incidence in Controls		
	Squamous Cell Papilloma[b]	Squamous Cell Carcinoma	Squamous Cell Papilloma or Carcinoma[b]
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	0/51	0/51	0/51
Benzyl Acetate	0/50	0/50	0/50
Butyl Benzyl Phthalate	0/50	0/50	0/50
C.I. Pigment Red 23	0/50	0/50	0/50
C.I. Pigment Red 3	1/50	0/50	1/50
o-Nitroanisole	0/50	0/50	0/50
p-Nitrobenzoic Acid	1/50	0/50	1/50
Polysorbate 80	1/50	0/50	1/50
Overall Historical Incidence			
Total	10/1,304 (0.8%)	0/1,304 (0%)	10/1,304 (0.8%)
Standard deviation	1.3%		1.3%
Range	0%-4%		0%-4%

[a] Data as of 12 May 1995. Includes data for oral mucosa, tongue, pharynx, and tooth.
[b] Includes data for papilloma.

TABLE A4d
Historical Incidence of Testicular Adenoma in Untreated Male F344/N Rats [a]

Study	Incidence in Controls
Historical Incidence at Southern Research Institute	
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	49/51
Benzyl Acetate	47/50
Butyl Benzyl Phthalate	44/50
C.I. Pigment Red 23	48/50
C.I. Pigment Red 3	47/50
o-Nitroanisole	48/50
p-Nitrobenzoic Acid	44/50
Polysorbate 80	39/49
Overall Historical Incidence	
Total	1,169/1,302 (89.8%)
Standard deviation	5.9%
Range	74%-98%

[a] Data as of 12 May 1995

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Disposition Summary				
Animals initially in study	60	60	60	60
12-Month interim evaluation	10	9	9	6
Early deaths				
Moribund	29	29	41	49
Natural deaths	2	2	1	3
Other			1	
Survivors				
Terminal sacrifice	19	20	8	2
Animals examined microscopically	60	60	60	60
12-Month Interim Evaluation				
Alimentary System				
Intestine large, colon	(9)	(9)	(8)	(5)
Parasite metazoan	1 (11%)		1 (13%)	
Intestine large, rectum	(10)	(9)	(9)	(6)
Parasite metazoan	1 (10%)		2 (22%)	
Intestine large, cecum	(10)	(9)	(9)	(6)
Parasite metazoan		1 (11%)	1 (11%)	
Liver	(10)	(9)	(9)	(6)
Basophilic focus	1 (10%)	1 (11%)		1 (17%)
Clear cell focus			1 (11%)	1 (17%)
Eosinophilic focus		2 (22%)		
Granuloma	1 (10%)			
Hepatodiaphragmatic nodule	1 (10%)	1 (11%)		
Inflammation, subacute	1 (10%)	2 (22%)	2 (22%)	2 (33%)
Mixed cell focus				1 (17%)
Bile duct, hyperplasia	4 (40%)			1 (17%)
Bile duct, pigmentation		9 (100%)	9 (100%)	6 (100%)
Hepatocyte, cytologic alterations		8 (89%)	9 (100%)	6 (100%)
Hepatocyte, pigmentation		1 (11%)	8 (89%)	6 (100%)
Kupffer cell, pigmentation				5 (83%)
Mesentery	(1)		(1)	
Fat, necrosis	1 (100%)		1 (100%)	
Oral mucosa				(1)
Hyperplasia				1 (100%)
Pancreas	(10)	(9)	(9)	(6)
Atrophy		2 (22%)	4 (44%)	2 (33%)
Inflammation, chronic			1 (11%)	
Cardiovascular System				
Heart	(10)	(9)	(9)	(6)
Cardiomyopathy	5 (50%)	3 (33%)	2 (22%)	3 (50%)

[a] Number of animals examined microscopically at the site and the number of animals with lesion

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
12-Month Interim Evaluation (continued)				
Endocrine System				
Adrenal cortex	(10)	(9)	(9)	(6)
Accessory adrenal cortical nodule		1 (11%)	1 (11%)	1 (17%)
Hypertrophy, focal		1 (11%)		
Pituitary gland	(10)	(9)	(8)	(6)
Pars distalis, angiectasis	1 (10%)			
Pars distalis, cyst	1 (10%)	1 (11%)		
Pars distalis, hyperplasia, focal	1 (10%)			1 (17%)
Pars intermedia, angiectasis	1 (10%)			
Thyroid gland	(10)	(9)	(9)	(6)
Ultimobranchial cyst	1 (10%)		2 (22%)	
Genital System				
Epididymis	(10)	(9)	(9)	(6)
Atypia cellular		1 (11%)		
Hypospermia	1 (10%)			
Preputial gland	(10)	(9)	(9)	(6)
Inflammation, chronic	5 (50%)	3 (33%)	2 (22%)	2 (33%)
Prostate	(10)	(9)	(9)	(6)
Corpora amylacea		1 (11%)	1 (11%)	1 (17%)
Inflammation, suppurative	3 (30%)	7 (78%)	5 (56%)	4 (67%)
Testes	(10)	(9)	(9)	(6)
Interstitial cell, hyperplasia	7 (70%)	4 (44%)	3 (33%)	3 (50%)
Seminiferous tubule, atrophy	2 (20%)			1 (17%)
Hematopoietic System				
Lymph node	(2)	(2)	(4)	(2)
Mediastinal, hemorrhage	2 (100%)	2 (100%)	2 (50%)	1 (50%)
Mediastinal, hyperplasia, lymphoid			2 (50%)	
Mediastinal, pigmentation	2 (100%)	1 (50%)	1 (25%)	2 (100%)
Lymph node, mandibular	(10)	(8)	(9)	(6)
Hemorrhage	5 (50%)		2 (22%)	1 (17%)
Hyperplasia, lymphoid		1 (13%)		
Pigmentation	1 (10%)	1 (13%)	3 (33%)	1 (17%)
Lymph node, mesenteric	(10)	(9)	(9)	(6)
Ectasia	2 (20%)			1 (17%)
Hemorrhage			1 (11%)	
Respiratory System				
Lung	(10)	(9)	(9)	(6)
Hemorrhage				1 (17%)
Infiltration cellular, histiocyte	2 (20%)	2 (22%)	2 (22%)	1 (17%)
Inflammation, subacute		3 (33%)		3 (50%)
Alveolar epithelium, hyperplasia				1 (17%)
Nose	(10)	(9)	(9)	(6)
Mucosa, metaplasia, squamous		1 (11%)		

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
12-Month Interim Evaluation (continued)				
Special Senses System				
Eye				(3)
Cataract				1 (33%)
Hemorrhage				2 (67%)
Retina, degeneration				1 (33%)
Urinary System				
Kidney	(10)	(9)	(9)	(6)
Infarct			1 (11%)	
Mineralization	2 (20%)			
Nephropathy	10 (100%)	9 (100%)	9 (100%)	6 (100%)
Renal tubule, pigmentation		7 (78%)	9 (100%)	6 (100%)
Systems Examined With No Lesions Observed				
General Body System				
Integumentary System				
Musculoskeletal System				
Nervous System				
2-Year Study				
Alimentary System				
Intestine large, colon	(50)	(50)	(50)	(54)
Edema			1 (2%)	1 (2%)
Parasite metazoan	4 (8%)	4 (8%)	8 (16%)	3 (6%)
Intestine large, rectum	(50)	(49)	(51)	(54)
Parasite metazoan	9 (18%)	1 (2%)	3 (6%)	8 (15%)
Intestine large, cecum	(50)	(50)	(50)	(54)
Edema	1 (2%)	4 (8%)	8 (16%)	12 (22%)
Parasite metazoan	2 (4%)	1 (2%)	1 (2%)	3 (6%)
Intestine small, duodenum	(50)	(51)	(51)	(54)
Erosion			1 (2%)	
Epithelium, hyperplasia	1 (2%)	4 (8%)	22 (43%)	21 (39%)
Intestine small, jejunum	(50)	(50)	(51)	(54)
Cyst				1 (2%)
Inflammation, chronic				2 (4%)
Ulcer				1 (2%)
Epithelium, hyperplasia		3 (6%)	10 (20%)	12 (22%)
Intestine small, ileum	(49)	(50)	(50)	(54)
Epithelium, hyperplasia		3 (6%)	10 (20%)	8 (15%)
Liver	(50)	(51)	(51)	(54)
Angiectasis	6 (12%)	6 (12%)	11 (22%)	12 (22%)
Basophilic focus	14 (28%)	8 (16%)	6 (12%)	7 (13%)
Clear cell focus	9 (18%)	15 (29%)	15 (29%)	18 (33%)
Congestion				3 (6%)
Cyst			2 (4%)	
Degeneration, cystic	15 (30%)	19 (37%)	10 (20%)	13 (24%)
Developmental malformation			2 (4%)	
Eosinophilic focus	7 (14%)	5 (10%)	14 (27%)	12 (22%)
Granuloma	2 (4%)	2 (4%)	1 (2%)	

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Alimentary System (continued)				
Liver (continued)	(50)	(51)	(51)	(54)
Hematopoietic cell proliferation			5 (10%)	1 (2%)
Hepatodiaphragmatic nodule	1 (2%)	1 (2%)		3 (6%)
Inflammation, subacute			9 (18%)	3 (6%)
Mixed cell focus	1 (2%)	10 (20%)	9 (18%)	10 (19%)
Necrosis, focal	2 (4%)	11 (22%)	3 (6%)	3 (6%)
Thrombosis	2 (4%)	4 (8%)	1 (2%)	6 (11%)
Bile duct, hyperplasia	49 (98%)	26 (51%)	18 (35%)	32 (59%)
Bile duct, pigmentation		38 (75%)	51 (100%)	54 (100%)
Centrilobular, atrophy	14 (28%)	17 (33%)	8 (16%)	15 (28%)
Centrilobular, necrosis	1 (2%)			
Hepatocyte, cytologic alterations		20 (39%)	44 (86%)	42 (78%)
Hepatocyte, pigmentation		22 (43%)	45 (88%)	51 (94%)
Hepatocyte, vacuolization cytoplasmic	5 (10%)		1 (2%)	2 (4%)
Kupffer cell, pigmentation	7 (14%)	15 (29%)	23 (45%)	26 (48%)
Mesentery	(11)	(12)	(9)	(13)
Accessory spleen				1 (8%)
Angiectasis		1 (8%)		1 (8%)
Cyst				1 (8%)
Fat, necrosis	11 (100%)	12 (100%)	9 (100%)	12 (92%)
Pancreas	(50)	(51)	(51)	(54)
Atrophy	26 (52%)	30 (59%)	23 (45%)	22 (41%)
Basophilic focus	1 (2%)	3 (6%)	2 (4%)	1 (2%)
Edema	3 (6%)	10 (20%)	16 (31%)	22 (41%)
Metaplasia	1 (2%)			1 (2%)
Thrombosis				2 (4%)
Acinar cell, cytoplasmic alteration		5 (10%)	11 (22%)	8 (15%)
Acinar cell, hyperplasia, focal	6 (12%)	3 (6%)	6 (12%)	9 (17%)
Salivary glands	(50)	(51)	(50)	(54)
Atrophy	1 (2%)	1 (2%)	5 (10%)	7 (13%)
Basophilic focus			1 (2%	
Edema		1 (2%)	6 (12%)	21 (39%)
Inflammation, chronic				1 (2%)
Stomach, forestomach	(50)	(51)	(51)	(54)
Edema	5 (10%)	10 (20%)	15 (29%)	13 (24%)
Perforation				1 (2%)
Ulcer	5 (10%)	3 (6%)	5 (10%)	5 (9%)
Mucosa, hyperplasia	4 (8%)	13 (25%)	19 (37%)	21 (39%)
Stomach, glandular	(50)	(51)	(51)	(54)
Edema	7 (14%)	2 (4%)	7 (14%)	8 (15%)
Erosion	2 (4%)		1 (2%)	1 (2%)
Ulcer	1 (2%)			
Tongue	(1)		(1)	(4)
Hyperplasia				1 (25%)
Epithelium, hyperplasia				1 (25%)
Tooth		(1)	(3)	
Necrosis		1 (100%)	2 (67%)	

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Cardiovascular System				
Blood vessel	(50)	(51)	(51)	(53)
Hypertrophy	1 (2%)	3 (6%)		3 (6%)
Inflammation, subacute	1 (2%)	2 (4%)		3 (6%)
Mineralization				1 (2%)
Necrosis				1 (2%)
Thrombosis		3 (6%)	2 (4%)	3 (6%)
Heart	(50)	(51)	(51)	(54)
Cardiomyopathy	39 (78%)	32 (63%)	33 (65%)	27 (50%)
Mineralization				2 (4%)
Thrombosis			1 (2%)	
Endocrine System				
Adrenal cortex	(50)	(51)	(51)	(54)
Accessory adrenal cortical nodule	10 (20%)	20 (39%)	12 (24%)	13 (24%)
Angiectasis			1 (2%)	1 (2%)
Degeneration, fatty	11 (22%)	5 (10%)	7 (14%)	6 (11%)
Hyperplasia, diffuse				2 (4%)
Hyperplasia, focal		1 (2%)	2 (4%)	2 (4%)
Hypertrophy, focal	5 (10%)	1 (2%)	2 (4%)	1 (2%)
Necrosis	1 (2%)	1 (2%)		
Adrenal medulla	(50)	(50)	(51)	(54)
Hyperplasia	13 (26%)	20 (40%)	18 (35%)	19 (35%)
Islets, pancreatic	(50)	(51)	(51)	(54)
Hyperplasia	1 (2%)			
Parathyroid gland	(47)	(47)	(48)	(52)
Cyst			1 (2%)	
Hyperplasia	3 (6%)	9 (19%)	15 (31%)	17 (33%)
Pituitary gland	(50)	(50)	(50)	(52)
Pars distalis, angiectasis	6 (12%)	2 (4%)	6 (12%)	3 (6%)
Pars distalis, cyst	3 (6%)	5 (10%)	5 (10%)	7 (13%)
Pars distalis, hyperplasia, focal	11 (22%)	7 (14%)	6 (12%)	9 (17%)
Pars intermedia, angiectasis		1 (2%)	1 (2%)	
Pars intermedia, cyst		2 (4%)	3 (6%)	3 (6%)
Pars nervosa, developmental malformation			1 (2%)	
Thyroid gland	(50)	(51)	(50)	(54)
Ultimobranchial cyst	1 (2%)	4 (8%)		4 (7%)
C-cell, hyperplasia	2 (4%)	6 (12%)	1 (2%)	6 (11%)
Follicle, cyst	1 (2%)	2 (4%)	6 (12%)	7 (13%)
General Body System				
None				
Genital System				
Epididymis	(49)	(51)	(51)	(54)
Atypia cellular	19 (39%)	27 (53%)	27 (53%)	23 (43%)
Edema	1 (2%)	3 (6%)	5 (10%)	14 (26%)
Granuloma sperm			1 (2%)	1 (2%)
Hypospermia	25 (51%)	37 (73%)	38 (75%)	29 (54%)

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Genital System (continued)				
Preputial gland	(49)	(50)	(51)	(54)
Cyst	3 (6%)	4 (8%)	1 (2%)	
Hyperplasia	2 (4%)		1 (2%)	2 (4%)
Inflammation, chronic	10 (20%)	6 (12%)	10 (20%)	4 (7%)
Inflammation, suppurative	8 (16%)	5 (10%)	7 (14%)	1 (2%)
Prostate	(50)	(51)	(51)	(53)
Corpora amylacea	18 (36%)	28 (55%)	24 (47%)	17 (32%)
Edema	1 (2%)	2 (4%)	2 (4%)	12 (23%)
Inflammation, suppurative	34 (68%)	22 (43%)	31 (61%)	34 (64%)
Epithelium, hyperplasia	6 (12%)	5 (10%)	3 (6%)	1 (2%)
Seminal vesicle	(50)	(51)	(51)	(54)
Edema		1 (2%)		1 (2%)
Testes	(49)	(51)	(51)	(54)
Interstitial cell, hyperplasia	6 (12%)	4 (8%)	4 (8%)	3 (6%)
Seminiferous tubule, atrophy	2 (4%)	4 (8%)	2 (4%)	5 (9%)
Hematopoietic System				
Bone marrow	(50)	(51)	(51)	(54)
Hemorrhage			5 (10%)	3 (6%)
Hyperplasia	3 (6%)	1 (2%)	4 (8%)	4 (7%)
Myelofibrosis	3 (6%)	3 (6%)	1 (2%)	4 (7%)
Lymph node	(20)	(23)	(26)	(45)
Hemorrhage			2 (8%)	4 (9%)
Hyperplasia, lymphoid			2 (8%)	4 (9%)
Pigmentation			1 (4%)	2 (4%)
Axillary, ectasia			1 (4%)	
Axillary, hemorrhage			1 (4%)	
Axillary, hyperplasia, lymphoid			1 (4%)	
Axillary, pigmentation			1 (4%)	
Deep cervical, hemorrhage			2 (8%)	
Deep cervical, hyperplasia, lymphoid			1 (4%)	
Deep cervical, pigmentation	2 (10%)		4 (15%)	1 (2%)
Iliac, ectasia	1 (5%)			
Iliac, hemorrhage				1 (2%)
Iliac, hyperplasia, lymphoid				1 (2%)
Iliac, pigmentation			1 (4%)	1 (2%)
Inguinal, hemorrhage			2 (8%)	2 (4%)
Inguinal, hyperplasia, lymphoid			2 (8%)	2 (4%)
Mediastinal, ectasia			3 (12%)	1 (2%)
Mediastinal, hemorrhage		3 (13%)	7 (27%)	16 (36%)
Mediastinal, hyperplasia, lymphoid			9 (35%)	17 (38%)
Mediastinal, pigmentation	8 (40%)	10 (43%)	14 (54%)	21 (47%)
Pancreatic, ectasia	2 (10%)			
Pancreatic, hemorrhage		1 (4%)	1 (4%)	
Pancreatic, hyperplasia, lymphoid			1 (4%)	6 (13%)
Pancreatic, pigmentation	1 (5%)	3 (13%)	3 (12%)	12 (27%)
Renal, ectasia				1 (2%)
Renal, hemorrhage			1 (4%)	2 (4%)
Renal, hyperplasia, lymphoid			1 (4%)	4 (9%)
Renal, pigmentation	3 (15%)		1 (4%)	11 (24%)

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Hematopoietic System (continued)				
Lymph node, mandibular	(50)	(51)	(50)	(53)
Ectasia	7 (14%)	8 (16%)	9 (18%)	3 (6%)
Hemorrhage	6 (12%)	4 (8%)	8 (16%)	11 (21%)
Hyperplasia, lymphoid	8 (16%)	12 (24%)	22 (44%)	25 (47%)
Pigmentation	5 (10%)	5 (10%)	4 (8%)	7 (13%)
Lymph node, mesenteric	(50)	(50)	(51)	(54)
Ectasia	6 (12%)	1 (2%)	2 (4%)	2 (4%)
Hemorrhage			9 (18%)	7 (13%)
Hyperplasia, lymphoid	3 (6%)	2 (4%)	10 (20%)	14 (26%)
Pigmentation			1 (2%)	
Spleen	(50)	(51)	(51)	(54)
Congestion		1 (2%)	3 (6%)	1 (2%)
Fibrosis	15 (30%)	16 (31%)	19 (37%)	17 (31%)
Hematopoietic cell proliferation	15 (30%)	12 (24%)	16 (31%)	16 (30%)
Metaplasia, lipocyte				2 (4%)
Necrosis		1 (2%)		
Pigmentation	9 (18%)	12 (24%)	14 (27%)	11 (20%)
Lymphoid follicle, hyperplasia				1 (2%)
Thymus	(48)	(49)	(49)	(53)
Hyperplasia				1 (2%)
Integumentary System				
Mammary gland	(48)	(48)	(49)	(53)
Hyperplasia	23 (48%)	11 (23%)	12 (24%)	7 (13%)
Skin	(50)	(50)	(51)	(54)
Cyst epithelial inclusion	3 (6%)	1 (2%)	1 (2%)	
Hyperkeratosis	4 (8%)	3 (6%)	3 (6%)	8 (15%)
Ulcer	2 (4%)			1 (2%)
Epidermis, hyperplasia	5 (10%)	5 (10%)	5 (10%)	9 (17%)
Subcutaneous tissue, edema		1 (2%)	4 (8%)	16 (30%)
Musculoskeletal System				
Bone	(50)	(51)	(51)	(54)
Fibrous osteodystrophy	2 (4%)	8 (16%)	18 (35%)	14 (26%)
Hyperostosis	1 (2%)		1 (2%)	
Skeletal muscle	(1)	(2)	(5)	(9)
Edema		1 (50%)	4 (80%)	9 (100%)
Nervous System				
Brain	(50)	(51)	(51)	(54)
Developmental malformation	8 (16%)	4 (8%)	4 (8%)	3 (6%)
Hemorrhage			2 (4%)	1 (2%)
Hydrocephalus	2 (4%)		1 (2%)	1 (2%)
Thrombosis				1 (2%)

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Respiratory System				
Lung	(50)	(51)	(51)	(54)
Congestion				2 (4%)
Cyst			1 (2%)	
Edema	1 (2%)	6 (12%)	10 (20%)	19 (35%)
Hemorrhage	2 (4%)	2 (4%)		2 (4%)
Infiltration cellular, histiocyte	17 (34%)	13 (25%)	18 (35%)	14 (26%)
Inflammation, subacute	2 (4%)	4 (8%)	2 (4%)	1 (2%)
Metaplasia, osseous				2 (4%)
Alveolar epithelium, hyperplasia	4 (8%)	6 (12%)	2 (4%)	2 (4%)
Nose	(50)	(51)	(51)	(54)
Exudate	8 (16%)	7 (14%)	10 (20%)	10 (19%)
Foreign body	2 (4%)		2 (4%)	6 (11%)
Mucosa, hyperplasia	5 (10%)	8 (16%)	9 (18%)	8 (15%)
Mucosa, metaplasia, squamous	7 (14%)	3 (6%)	7 (14%)	8 (15%)
Special Senses System				
Eye	(1)			(1)
Cataract	1 (100%)			
Congestion				1 (100%)
Retina, degeneration	1 (100%)			
Urinary System				
Kidney	(50)	(51)	(51)	(54)
Cyst	1 (2%)	6 (12%)	7 (14%)	8 (15%)
Hydronephrosis		2 (4%)		1 (2%)
Inflammation, suppurative		1 (2%)	5 (10%)	4 (7%)
Mineralization	4 (8%)			2 (4%)
Nephropathy	50 (100%)	51 (100%)	51 (100%)	54 (100%)
Pelvis, hemorrhage				1 (2%)
Renal tubule, hyperplasia			4 (8%)	3 (6%)
Renal tubule, pigmentation	18 (36%)	43 (84%)	47 (92%)	54 (100%)
Transitional epithelium, hyperplasia	11 (22%)	23 (45%)	29 (57%)	34 (63%)
Urinary bladder	(50)	(51)	(51)	(54)
Hemorrhage				1 (2%)
Transitional epithelium, hyperplasia	1 (2%)	1 (2%)	1 (2%)	1 (2%)

APPENDIX B
SUMMARY OF LESIONS IN FEMALE RATS IN THE 2-YEAR FEED STUDY OF D&C YELLOW NO. 11

TABLE B1 Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 101

TABLE B2 Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 106

TABLE B3 Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 124

TABLE B4a Historical Incidence of Hepatocellular Neoplasms in Untreated Female F344/N Rats 129

TABLE B4b Historical Incidence of Renal Tubule Neoplasms in Untreated Female F344/N Rats 129

TABLE B4c Historical Incidence of Oral Cavity Neoplasms in Untreated Female F344/N Rats 130

TABLE B4d Historical Incidence of Clitoral Gland Neoplasms in Untreated Female F344/N Rats 130

TABLE B5 Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 131

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Disposition Summary				
Animals initially in study	60	60	60	60
12-Month interim evaluation	10	9	10	9
Early deaths				
Moribund	25	23	12	25
Natural deaths	3	2	1	3
Survivors				
Terminal sacrifice	22	26	37	23
Animals examined microscopically	60	60	60	60
12-Month Interim Evaluation				
Endocrine System				
Pituitary gland	(10)	(9)	(10)	(9)
Pars distalis, adenoma				2 (22%)
Genital System				
Clitoral gland	(10)	(9)	(10)	(9)
Carcinoma		1 (11%)		1 (11%)
Uterus	(10)	(9)	(10)	(9)
Polyp stromal	2 (20%)	2 (22%)	3 (30%)	
Hematopoietic System				
Spleen	(10)	(9)	(10)	(9)
Fibrous histiocytoma				1 (11%)
Integumentary System				
Mammary gland	(10)	(9)	(10)	(9)
Carcinoma	1 (10%)			1 (11%)
Skin	(10)	(9)	(10)	(9)
Subcutaneous tissue, schwannoma malignant	1 (10%)			1 (11%)
Systems Examined With No Neoplasms Observed				
Alimentary System				
Cardiovascular System				
General Body System				
Musculoskeletal System				
Nervous System				
Respiratory System				
Special Senses System				
Urinary System				

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study				
Alimentary System				
Intestine large, colon	(50)	(50)	(50)	(51)
Carcinoma		1 (2%)	1 (2%)	
Intestine large, rectum	(49)	(50)	(49)	(50)
Histiocytic sarcoma				1 (2%)
Intestine large, cecum	(50)	(51)	(49)	(51)
Intestine small, jejunum	(49)	(51)	(50)	(51)
Leiomyosarcoma	1 (2%)			
Intestine small, ileum	(48)	(51)	(49)	(49)
Liver	(50)	(51)	(50)	(51)
Cholangiocarcinoma	1 (2%)			1 (2%)
Hepatocellular carcinoma				1 (2%)
Hepatocellular adenoma		1 (2%)	5 (10%)	4 (8%)
Hepatocellular adenoma, multiple		1 (2%)		
Osteosarcoma, metastatic, bone		1 (2%)		
Mesentery	(11)	(18)	(8)	(12)
Cholangiocarcinoma, metastatic, liver				1 (8%)
Osteosarcoma, metastatic, bone		1 (6%)		
Oral mucosa	(1)	(1)		(1)
Squamous cell carcinoma				1 (100%)
Squamous cell papilloma	1 (100%)	1 (100%)		
Pancreas	(50)	(51)	(50)	(51)
Osteosarcoma, metastatic, bone		1 (2%)		
Salivary glands	(50)	(46)	(50)	(50)
Stomach, forestomach	(50)	(51)	(50)	(51)
Squamous cell carcinoma		1 (2%)		
Squamous cell papilloma		2 (4%)		2 (4%)
Stomach, glandular	(50)	(51)	(50)	(51)
Tongue				(1)
Squamous cell carcinoma				1 (100%)
Cardiovascular System				
Heart	(50)	(51)	(50)	(51)
Osteosarcoma, metastatic, bone				1 (2%)
Schwannoma malignant				1 (2%)
Endocrine System				
Adrenal cortex	(50)	(51)	(50)	(51)
Adenoma		1 (2%)		
Adrenal medulla	(48)	(51)	(50)	(51)
Pheochromocytoma malignant			1 (2%)	1 (2%)
Pheochromocytoma complex	1 (2%)			
Pheochromocytoma benign	2 (4%)	1 (2%)	2 (4%)	1 (2%)
Islets, pancreatic	(50)	(51)	(49)	(51)
Adenoma	1 (2%)	1 (2%)	1 (2%)	
Parathyroid gland	(48)	(48)	(48)	(50)
Adenoma			1 (2%)	1 (2%)
Pituitary gland	(50)	(51)	(50)	(51)
Pars distalis, adenoma	23 (46%)	23 (45%)	18 (36%)	20 (39%)
Pars distalis, adenoma, multiple	1 (2%)			
Pars distalis, carcinoma				2 (4%)

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Endocrine System (continued)				
Thyroid gland	(50)	(51)	(50)	(51)
Bilateral, follicular cell, carcinoma				1 (2%)
C-cell, adenoma	2 (4%)	2 (4%)	4 (8%)	5 (10%)
C-cell, adenoma, multiple				1 (2%)
C-cell, carcinoma		1 (2%)	1 (2%)	
Follicular cell, adenoma			1 (2%)	2 (4%)
Follicular cell, carcinoma		1 (2%)		
General Body System				
None				
Genital System				
Clitoral gland	(49)	(50)	(49)	(51)
Adenoma	11 (22%)	4 (8%)	5 (10%)	4 (8%)
Carcinoma	5 (10%)	2 (4%)	6 (12%)	2 (4%)
Bilateral, carcinoma	1 (2%)			
Ovary	(50)	(51)	(50)	(51)
Granulosa cell tumor malignant			1 (2%)	
Granulosa cell tumor benign			1 (2%)	
Osteosarcoma, metastatic, bone		1 (2%)		
Uterus	(50)	(51)	(50)	(51)
Adenoma				1 (2%)
Carcinoma	1 (2%)			
Cholangiocarcinoma, metastatic, liver				1 (2%)
Polyp stromal	11 (22%)	11 (22%)	7 (14%)	6 (12%)
Polyp stromal, multiple	1 (2%)			
Schwannoma malignant	1 (2%)	1 (2%)		
Hematopoietic System				
Bone marrow	(50)	(51)	(50)	(51)
Lymph node	(9)	(11)	(11)	(15)
Lymph node, mandibular	(50)	(51)	(50)	(49)
Lymph node, mesenteric	(50)	(51)	(50)	(51)
Lymph node, mediastinal		(1)		(1)
Cholangiocarcinoma, metastatic, liver				1 (100%)
Spleen	(50)	(50)	(50)	(51)
Thymus	(50)	(49)	(49)	(49)
Integumentary System				
Mammary gland	(50)	(51)	(50)	(51)
Adenoma	2 (4%)			
Carcinoma	4 (8%)	5 (10%)	2 (4%)	3 (6%)
Carcinoma, multiple			1 (2%)	
Fibroadenoma	14 (28%)	13 (25%)	18 (36%)	17 (33%)
Fibroadenoma, multiple	7 (14%)	9 (18%)	5 (10%)	9 (18%)
Histiocytic sarcoma				1 (2%)

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Integumentary System (continued)				
Skin	(49)	(51)	(50)	(51)
Basal cell carcinoma				1 (2%)
Histiocytic sarcoma				1 (2%)
Squamous cell carcinoma			2 (4%)	
Squamous cell papilloma		1 (2%)		1 (2%)
Subcutaneous tissue, fibroma	1 (2%)			
Musculoskeletal System				
Bone	(50)	(51)	(50)	(51)
Osteosarcoma	1 (2%)	1 (2%)		1 (2%)
Skeletal muscle		(2)		(1)
Osteosarcoma, metastatic, bone		1 (50%)		
Rhabdomyosarcoma				1 (100%)
Nervous System				
Brain	(50)	(51)	(50)	(51)
Astrocytoma malignant		1 (2%)		
Carcinoma, metastatic, pituitary gland				2 (4%)
Respiratory System				
Lung	(50)	(51)	(50)	(51)
Alveolar/bronchiolar adenoma	1 (2%)		2 (4%)	1 (2%)
Alveolar/bronchiolar adenoma, multiple		1 (2%)		
Alveolar/bronchiolar carcinoma				2 (4%)
Carcinoma, metastatic, uterus	1 (2%)			
Cholangiocarcinoma, metastatic, liver				1 (2%)
Osteosarcoma, metastatic, bone				1 (2%)
Special Senses System				
Zymbal's gland				(1)
Carcinoma				1 (100%)
Urinary System				
Kidney	(50)	(51)	(50)	(51)
Sarcoma			1 (2%)	
Renal tubule, carcinoma			1 (2%)	
Transitional epithelium, carcinoma	1 (2%)			
Transitional epithelium, hemangioma			1 (2%)	
Urinary bladder	(50)	(51)	(50)	(51)
Papilloma				1 (2%)
Systemic Lesions				
Multiple organs[b]	(50)	(51)	(50)	(51)
Histiocytic sarcoma				1 (2%)
Leukemia mononuclear	16 (32%)	21 (41%)	19 (38%)	16 (31%)
Lymphoma malignant		1 (2%)		

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Neoplasm Summary				
Total animals with primary neoplasms[c]				
12-Month interim evaluation	4	3	3	4
2-Year study	48	50	50	49
Total primary neoplasms				
12-Month interim evaluation	4	3	3	6
2-Year study	111	108	107	113
Total animals with benign neoplasms				
12-Month interim evaluation	2	2	3	2
2-Year study	42	44	41	41
Total benign neoplasms				
12-Month interim evaluation	2	2	3	2
2-Year study	78	72	71	76
Total animals with malignant neoplasms				
12-Month interim evaluation	2	1		2
2-Year study	30	29	30	28
Total malignant neoplasms				
12-Month interim evaluation	2	1		4
2-Year study	33	36	36	37
Total animals with metastatic neoplasms				
2-Year study	1	1		4
Total metastatic neoplasms				
2-Year study	1	5		8

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm

Number of Days on Study	481	537	570	572	572	589	589	602	607	629	650	651	656	660	671	684	699	700	701	712	719	722	729	730	730
Carcass ID Number	291	296	286	263	271	248	288	265	257	298	249	281	252	254	245	258	262	294	244	255	247	246	250	251	273
Alimentary System																									
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+
Leiomyosarcoma																			X						
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cholangiocarcinoma																X									
Mesentery				+			+		+		+					+						+			
Oral mucosa																									
Squamous cell papilloma																									
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tooth					+																				
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma complex																				X					
Pheochromocytoma benign																					X				
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																		X							
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma		X			X	X				X		X			X		X		X	X	X		X	X	X
Pars distalis, adenoma, multiple									X																
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-cell, adenoma																									
General Body System																									
None																									
Genital System																									
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+
Adenoma										X						X							X		
Carcinoma														X											X
Bilateral, carcinoma																									
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

+: Tissue examined microscopically
A: Autolysis precludes examination
M: Missing tissue
I: Insufficient tissue
X: Lesion present
Blank: Not examined

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	734	734	734	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	
Carcass ID Number	279	284	293	241	243	256	259	261	264	266	268	269	270	274	275	276	277	280	282	285	287	290	295	297	300	Total Tissues/ Tumors
Alimentary System																										
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	49
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Leiomyosarcoma																										1
Intestine small, ileum	+	+	+	+	+	+	+	+	I	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Cholangiocarcinoma																										1
Mesentery	+	+			+										+					+						11
Oral mucosa																		+								1
Squamous cell papilloma																		X								1
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Tooth																										1
Cardiovascular System																										
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Endocrine System																										
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adrenal medulla	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Pheochromocytoma complex																										1
Pheochromocytoma benign																	X									2
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adenoma																										1
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	48
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pars distalis, adenoma			X	X			X	X	X	X	X				X	X									X	23
Pars distalis, adenoma, multiple																										1
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
C-cell, adenoma					X			X																		2
General Body System																										
None																										
Genital System																										
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Adenoma	X				X	X			X			X		X		X						X				11
Carcinoma								X											X		X					5
Bilateral, carcinoma																								X		1
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	481	537	570	572	572	589	589	602	607	629	650	651	656	660	671	684	699	700	701	712	719	722	729	730	730
Carcass ID Number	291	296	286	263	271	248	288	265	257	298	249	281	252	254	245	258	262	294	244	255	247	246	250	251	273
Genital System (continued)																									
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Polyp stromal			X					X		X		X				X			X		X		X		
Polyp stromal, multiple																									
Schwannoma malignant			X																						
Vagina								M	M	M								M				M			
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node	+						+	+								+		+				+			
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																		X		X					
Carcinoma	X			X																	X		X		
Fibroadenoma											X				X			X					X		
Fibroadenoma, multiple						X								X					X					X	X
Skin	+	I	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Subcutaneous tissue, fibroma																	X								
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma													X												
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Peripheral nerve												+						+							
Spinal cord												+						+							
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma																									
Carcinoma, metastatic, uterus																									
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Special Senses System																									
Eye																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Transitional epithelium, carcinoma									X																
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear							X	X			X					X		X			X	X			X

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 0 ppm (continued)

Number of Days on Study	734	734	734	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	742	
Carcass ID Number	279	284	293	441	443	556	559	561	564	566	568	569	570	574	575	576	577	580	582	585	587	590	595	597	530	Total Tissues/ Tumors
Genital System (continued)																										
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma																		X								1
Polyp stromal		X	X				X																			11
Polyp stromal, multiple													X													1
Schwannoma malignant																										1
Vagina																		M				+				1
Hematopoietic System																										
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node	+																	+		+						9
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Integumentary System																										
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adenoma																										2
Carcinoma																										4
Fibroadenoma			X	X		X			X				X	X			X		X			X			X	14
Fibroadenoma, multiple		X																			X					7
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Subcutaneous tissue, fibroma																										1
Musculoskeletal System																										
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Osteosarcoma																										1
Nervous System																										
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Peripheral nerve																										2
Spinal cord														+												3
Respiratory System																										
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Alveolar/bronchiolar adenoma																		X								1
Carcinoma, metastatic, uterus																		X								1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Special Senses System																										
Eye														+												1
Urinary System																										
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Transitional epithelium, carcinoma																										1
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Systemic Lesions																										
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Leukemia mononuclear	X	X			X							X	X	X		X	X									16

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm

Number of Days on Study	308	462	546	548	600	601	622	622	628	629	631	631	642	645	656	672	688	688	693	695	705	712	716	726	734
Carcass ID Number	322	359	358	315	354	336	303	313	326	320	314	340	335	324	349	310	316	318	346	341	329	344	343	321	301
Alimentary System																									
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+
Intestine large, colon	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	I
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hepatocellular adenoma																									
Hepatocellular adenoma, multiple														X											
Osteosarcoma, metastatic, bone		X																							
Mesentery		+	+					+	+	+	+		+	+			+		+	+				+	+
Osteosarcoma, metastatic, bone		X																							
Oral mucosa									+																
Squamous cell papilloma									X																
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma, metastatic, bone		X																							
Salivary glands	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Squamous cell carcinoma																									
Squamous cell papilloma																									
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																									
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma benign											X														
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																									
Parathyroid gland	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma						X					X					X	X		X		X	X	X		X
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-cell, adenoma																									
C-cell, carcinoma																									
Follicular cell, carcinoma																									
General Body System																									
None																									
Genital System																									
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																									
Carcinoma				X																		X			
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma, metastatic, bone		X																							

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm (continued)

Number of Days on Study	740	740	740	740	740	740	740	740	740	740	740	740	740	740	740	741	741	741	741	741	741	741	741	741	741	741	
Carcass ID Number	302	304	306	308	309	311	317	323	325	328	330	331	333	334	338	339	342	345	347	350	351	352	353	355	356	360	Total Tissues/ Tumors
Alimentary System																											
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	I	+	+	+	+	48
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma											X																1
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Hepatocellular adenoma				X																							1
Hepatocellular adenoma, multiple																											1
Osteosarcoma, metastatic, bone																											1
Mesentery						+			+							+							+			+	18
Osteosarcoma, metastatic, bone																											1
Oral mucosa																											1
Squamous cell papilloma																											1
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Osteosarcoma, metastatic, bone																											1
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+		+				46
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Squamous cell carcinoma																	X										1
Squamous cell papilloma															X									X			2
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Cardiovascular System																											
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Endocrine System																											
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenoma							X																				1
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Pheochromocytoma benign																											1
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenoma													X														1
Parathyroid gland	+	+	+	M	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Pars distalis, adenoma	X			X			X		X	X		X		X	X	X	X					X	X	X		X	23
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
C-cell, adenoma				X																				X			2
C-cell, carcinoma																X											1
Follicular cell, carcinoma		X																									1
General Body System																											
None																											
Genital System																											
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	50
Adenoma						X						X										X			X		4
Carcinoma																											2
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Osteosarcoma, metastatic, bone																											1

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm (continued)

Number of Days on Study	308	462	546	548	600	601	622	622	628	629	631	631	642	645	656	672	688	688	693	695	705	712	716	726	734
Carcass ID Number	322	359	358	315	354	336	303	313	326	320	314	340	335	324	349	310	316	318	346	341	329	344	343	321	301
Genital System (continued)																									
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Polyp stromal				X							X	X			X								X	X	X
Schwannoma malignant																							X		
Vagina									M	M	M											M	M		
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node	+	+			+	+			+						+							+	+	+	
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mediastinal	+																								
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+
Thymus	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma									X			X													
Fibroadenoma				X			X	X								X	X							X	X
Fibroadenoma, multiple					X					X			X					X	X						
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Squamous cell papilloma																									
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma		X																							
Skeletal muscle		+																							
Osteosarcoma, metastatic, bone		X																							
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Astrocytoma malignant																							X		
Peripheral nerve														+									+		
Spinal cord														+									+		
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma, multiple																									
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Special Senses System																									
Ear							+																+		
Eye							+																		
Harderian gland																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear	X		X			X				X	X	X	X	X	X	X	X			X		X		X	
Lymphoma malignant									X																

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 500 ppm (continued)

Number of Days on Study	740	740	740	740	740	740	740	740	740	740	740	740	740	740	740	741	741	741	741	741	741	741	741	741	741	741	
Carcass ID Number	302	304	306	308	309	311	317	323	325	328	330	331	333	334	338	339	342	345	347	350	351	352	353	355	356	360	Total Tissues/ Tumors
Genital System (continued)																											
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Polyp stromal			X					X						X			X										11
Schwannoma malignant																											1
Vagina																											
Hematopoietic System																											
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node											+													+			11
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node, mediastinal																											1
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Integumentary System																											
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Carcinoma									X						X	X											5
Fibroadenoma		X		X	X												X							X	X		13
Fibroadenoma, multiple								X										X	X				X				9
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Squamous cell papilloma																	X										1
Musculoskeletal System																											
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Osteosarcoma																											1
Skeletal muscle																				+							2
Osteosarcoma, metastatic, bone																											1
Nervous System																											
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Astrocytoma malignant																											1
Peripheral nerve																											2
Spinal cord																											2
Respiratory System																											
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Alveolar/bronchiolar adenoma, multiple												X															1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Special Senses System																											
Ear																											2
Eye					+																						2
Harderian gland					+																						1
Urinary System																											
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Systemic Lesions																											
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Leukemia mononuclear					X	X					X						X			X				X	X		21
Lymphoma malignant																											1

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm

Number of Days on Study	456	532	588	597	610	638	658	660	671	701	719	722	722	740	740	740	740	740	740	740	740	740	740	740	740
Carcass ID Number	414	378	409	370	415	403	372	376	401	363	396	397	417	361	362	365	367	368	369	374	375	377	379	380	382
Alimentary System																									
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Intestine large, rectum	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Hepatocellular adenoma																	X		X						
Mesentery	+	+	+	+																			+		
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma malignant									X																
Pheochromocytoma benign							X										X								
Islets, pancreatic	+	I	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																								X	
Parathyroid gland	+	+	M	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																									
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma					X	X	X	X				X				X							X		X
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
C-cell, adenoma												X					X								
C-cell, carcinoma						X																			
Follicular cell, adenoma																					X				
General Body System																									
None																									
Genital System																									
Clitoral gland	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma														X											
Carcinoma	X																		X					X	
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Granulosa cell tumor malignant																					X				
Granulosa cell tumor benign																									
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Polyp stromal						X	X																	X	
Vagina		M																							

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	740	740	740	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	
Carcass ID Number	383	384	385	386	387	389	390	392	393	394	395	398	399	400	402	404	405	406	407	410	411	413	416	419	420	Total Tissues/ Tumors
Alimentary System																										
Esophagus	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	49
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma												X														1
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Hepatocellular adenoma	X																			X					X	5
Mesentery																+	+				+					8
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Cardiovascular System																										
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Endocrine System																										
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pheochromocytoma malignant																										1
Pheochromocytoma benign																										2
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Adenoma																										1
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	48
Adenoma																						X				1
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Pars distalis, adenoma					X			X		X		X		X	X	X	X		X	X						18
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
C-cell, adenoma												X		X												4
C-cell, carcinoma																										1
Follicular cell, adenoma																										1
General Body System																										
None																										
Genital System																										
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Adenoma		X													X					X					X	5
Carcinoma			X									X											X			6
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Granulosa cell tumor malignant																										1
Granulosa cell tumor benign																					X					1
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Polyp stromal						X	X							X				X								7
Vagina																										

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	456	532	588	597	610	638	658	660	671	701	719	722	722	740	740	740	740	740	740	740	740	740	740	740	740
Carcass ID Number	414	378	409	370	415	403	372	376	401	363	396	397	417	361	362	365	367	368	369	374	375	377	379	380	382
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node	+	+	+	+		+				+		+			+			+							
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Thymus	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma																									
Carcinoma, multiple												X													
Fibroadenoma	X					X			X	X			X					X	X			X	X		
Fibroadenoma, multiple								X			X										X			X	
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Squamous cell carcinoma						X										X									
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma																			X						
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Special Senses System																									
None																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Sarcoma																									
Renal tubule, carcinoma																									
Transitional epithelium, hemangioma																					X				
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Leukemia mononuclear		X	X	X					X	X	X	X	X		X	X	X			X					

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 1,700 ppm (continued)

Number of Days on Study	740	740	740	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	741	
Carcass ID Number	383	384	385	386	387	389	390	392	393	394	395	398	399	400	402	404	405	406	407	410	411	413	416	419	420	Total Tissues/ Tumors
Hematopoietic System																										
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node																		+			+					11
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Integumentary System																										
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Carcinoma		X														X										2
Carcinoma, multiple																										1
Fibroadenoma				X		X			X			X				X	X			X	X			X		18
Fibroadenoma, multiple	X																									5
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Squamous cell carcinoma																										2
Musculoskeletal System																										
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Nervous System																										
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Respiratory System																										
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Alveolar/bronchiolar adenoma																	X									2
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Special Senses System																										
None																										
Urinary System																										
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Sarcoma																				X						1
Renal tubule, carcinoma																X										1
Transitional epithelium, hemangioma																										1
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Systemic Lesions																										
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Leukemia mononuclear	X						X	X			X		X						X					X		19

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm

Number of Days on Study	147	141	148	169	106	130	168	190	196	148	156	156	158	158	172	195	100	112	120	123	123	128	128	128	129
Carcass ID Number	473	442	480	337	556	559	662	711	446	422	421	477	444	453	550	424	661	665	443	425	428	436	440	449	470
Alimentary System																									
Esophagus	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	I	+	+	+
Histiocytic sarcoma																X									
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	M	+	+	+	+	+	+	+	+
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Cholangiocarcinoma													X												
Hepatocellular carcinoma																						X			
Hepatocellular adenoma																			X						
Mesentery							+					+	+							+	+	+			
Cholangiocarcinoma, metastatic, liver													X												
Oral mucosa																									
Squamous cell carcinoma																									
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salivary glands	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Squamous cell papilloma								X																	
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tongue																									
Squamous cell carcinoma																									
Cardiovascular System																									
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma, metastatic, bone						X																			
Schwannoma malignant																									
Endocrine System																									
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pheochromocytoma malignant																					X				
Pheochromocytoma benign																									
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																	X								
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Pars distalis, adenoma			X								X	X			X		X	X	X	X				X	X
Pars distalis, carcinoma																					X				
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Bilateral, follicular cell, carcinoma																									
C-cell, adenoma												X									X				
C-cell, adenoma, multiple																		X							
Follicular cell, adenoma																			X	X					
General Body System																									
None																									

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	729	733	734	740	740	740	740	740	740	740	740	740	740	740	740	740	740	740	741	741	741	741	741	741	741	741	
Carcass ID Number	476	432	433	426	429	430	431	435	438	439	445	448	451	454	455	457	458	463	464	466	467	468	474	475	478	479	Total Tissues/ Tumors
Alimentary System																											
Esophagus	+	+	+	+	+	M	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	47
Intestine large, colon	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine large, rectum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Histiocytic sarcoma																											1
Intestine large, cecum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, duodenum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	50
Intestine small, jejunum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Intestine small, ileum	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	49
Liver	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Cholangiocarcinoma																											1
Hepatocellular carcinoma																											1
Hepatocellular adenoma						X												X						X			4
Mesentery					+	+	+		+				+	+													12
Cholangiocarcinoma, metastatic, liver																											1
Oral mucosa																							+				1
Squamous cell carcinoma																							X				1
Pancreas	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Salivary glands		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	50
Stomach, forestomach	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Squamous cell papilloma														X													2
Stomach, glandular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Tongue		+																									1
Squamous cell carcinoma		X																									1
Cardiovascular System																											
Blood vessel	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Heart	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Osteosarcoma, metastatic, bone																											1
Schwannoma malignant	X																										1
Endocrine System																											
Adrenal cortex	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adrenal medulla	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Pheochromocytoma malignant																											1
Pheochromocytoma benign																								X			1
Islets, pancreatic	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Parathyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	50
Adenoma																											1
Pituitary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Pars distalis, adenoma	X	X			X		X					X			X				X	X	X					X	20
Pars distalis, carcinoma								X																			2
Thyroid gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Bilateral, follicular cell, carcinoma																										X	1
C-cell, adenoma						X																X			X		5
C-cell, adenoma, multiple																											1
Follicular cell, adenoma																											2
General Body System																											
None																											

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	147	141	148	169	106	130	168	190	196	148	156	156	158	158	172	195	100	112	120	123	123	128	128	128	129
Carcass ID Number	473	442	480	337	556	559	662	771	446	422	421	477	444	553	550	424	461	465	443	425	428	436	440	449	470
Genital System																									
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma											X										X				
Carcinoma																						X			
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Adenoma																						X			
Cholangiocarcinoma, metastatic, liver													X												
Polyp stromal										X						X		X							
Vagina							M					M						M	M						
Hematopoietic System																									
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node	+	+		+		+	+	+	+					+				+		+	+				+
Lymph node, mandibular	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lymph node, mediastinal													+												
Cholangiocarcinoma, metastatic, liver													X												
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Thymus	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Integumentary System																									
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma					X							X													
Fibroadenoma							X				X			X		X							X	X	
Fibroadenoma, multiple													X					X	X		X				X
Histiocytic sarcoma																X									
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Basal cell carcinoma																									
Histiocytic sarcoma																X									
Squamous cell papilloma																									
Musculoskeletal System																									
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Osteosarcoma						X																			
Skeletal muscle																					+				
Rhabdomyosarcoma																					X				
Nervous System																									
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Carcinoma, metastatic, pituitary gland												X													
Peripheral nerve																				+	+				
Spinal cord																				+	+				

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	729	733	734	740	740	740	740	740	740	740	740	740	740	740	740	740	740	740	741	741	741	741	741	741	741	741	
Carcass ID Number	476	432	433	426	429	430	431	435	438	439	445	448	451	454	455	457	458	463	464	466	467	468	474	475	478	479	Total Tissues/ Tumors
Genital System																											
Clitoral gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenoma							X				X																4
Carcinoma										X																	2
Ovary	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Uterus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Adenoma																											1
Cholangiocarcinoma, metastatic, liver																											1
Polyp stromal		X	X												X												6
Vagina		M																									
Hematopoietic System																											
Bone marrow	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node					+													+							+		15
Lymph node, mandibular	+	M	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	I	+	+	+	+	49
Lymph node, mesenteric	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Lymph node, mediastinal																											1
Cholangiocarcinoma, metastatic, liver																											1
Spleen	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Thymus	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	M	+	+	+	+	+	+	+	+	+	+	49
Integumentary System																											
Mammary gland	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Carcinoma	X																										3
Fibroadenoma	X	X					X					X	X	X	X	X				X	X			X			17
Fibroadenoma, multiple			X	X		X											X										9
Histiocytic sarcoma																											1
Skin	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Basal cell carcinoma																				X							1
Histiocytic sarcoma																											1
Squamous cell papilloma													X														1
Musculoskeletal System																											
Bone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Osteosarcoma																											1
Skeletal muscle																											1
Rhabdomyosarcoma																											1
Nervous System																											
Brain	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Carcinoma, metastatic, pituitary gland								X																			2
Peripheral nerve																											2
Spinal cord																											2

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	147	441	448	469	506	530	568	590	596	648	656	656	658	658	672	695	700	712	720	723	723	728	728	728	729
Carcass ID Number	473	442	480	437	556	559	562	571	446	422	421	477	444	453	450	424	461	465	443	425	428	436	440	449	470
Respiratory System																									
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Alveolar/bronchiolar adenoma																									
Alveolar/bronchiolar carcinoma								X																	X
Cholangiocarcinoma, metastatic, liver													X												
Osteosarcoma, metastatic, bone						X																			
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Special Senses System																									
Ear									+																
Lacrimal gland															+										
Zymbal's gland																									
Carcinoma																									
Urinary System																									
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Papilloma																									
Systemic Lesions																									
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Histiocytic sarcoma																X									
Leukemia mononuclear		X		X					X	X			X	X		X		X	X	X		X			X

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of D&C Yellow No. 11: 5,000 ppm (continued)

Number of Days on Study	729	733	734	740	740	740	740	740	740	740	740	740	740	740	740	740	740	740	741	741	741	741	741	741	741	741	
Carcass ID Number	476	432	433	426	429	430	431	435	438	439	445	448	451	454	455	457	458	463	464	466	467	468	474	475	478	479	Total Tissues/ Tumors
Respiratory System																											
Lung	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Alveolar/bronchiolar adenoma													X														1
Alveolar/bronchiolar carcinoma																											2
Cholangiocarcinoma, metastatic, liver																											1
Osteosarcoma, metastatic, bone																											1
Nose	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Trachea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Special Senses System																											
Ear																											1
Lacrimal gland																											1
Zymbal's gland																		+									1
Carcinoma																		X									1
Urinary System																											
Kidney	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Urinary bladder	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Papilloma																		X									1
Systemic Lesions																											
Multiple organs	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	51
Histiocytic sarcoma																											1
Leukemia mononuclear														X				X						X	X		16

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Adrenal Medulla: Benign, Complex, or Malignant Pheochromocytoma				
Overall rate[a]	3/48 (6%)	1/51 (2%)	3/50 (6%)	2/51 (4%)
Adjusted rate[b]	10.7%	2.4%	7.2%	7.3%
Terminal rate[c]	1/22 (5%)	0/26 (0%)	1/37 (3%)	1/23 (4%)
First incidence (days)	712	631	658	723
Life table test[d]	P=0.557N	P=0.300N	P=0.510N	P=0.463N
Logistic regression test[d]	P=0.563N	P=0.287N	P=0.639N	P=0.471N
Cochran-Armitage test[d]	P=0.556N			
Fisher exact test[d]		P=0.286N	P=0.641N	P=0.471N
Clitoral Gland: Adenoma				
Overall rate	11/49 (22%)	4/50 (8%)	5/49 (10%)	4/51 (8%)
Adjusted rate	40.2%	16.0%	13.5%	13.7%
Terminal rate	7/22 (32%)	4/25 (16%)	5/37 (14%)	2/23 (9%)
First incidence (days)	629	740 (T)	740 (T)	656
Life table test	P=0.100N	P=0.029N	P=0.009N	P=0.043N
Logistic regression test	P=0.086N	P=0.043N	P=0.035N	P=0.037N
Cochran-Armitage test	P=0.098N			
Fisher exact test		P=0.041N	P=0.085N	P=0.038N
Clitoral Gland: Carcinoma				
Overall rate	6/49 (12%)	2/50 (4%)	6/49 (12%)	2/51 (4%)
Adjusted rate	23.3%	5.3%	15.2%	7.5%
Terminal rate	4/22 (18%)	0/25 (0%)	5/37 (14%)	1/23 (4%)
First incidence (days)	660	548	456	728
Life table test	P=0.213N	P=0.123N	P=0.317N	P=0.129N
Logistic regression test	P=0.203N	P=0.131N	P=0.603N	P=0.116N
Cochran-Armitage test	P=0.199N			
Fisher exact test		P=0.128N	P=0.620N	P=0.122N
Clitoral Gland: Adenoma or Carcinoma				
Overall rate	17/49 (35%)	6/50 (12%)	11/49 (22%)	6/51 (12%)
Adjusted rate	58.9%	20.5%	28.5%	20.6%
Terminal rate	11/22 (50%)	4/25 (16%)	10/37 (27%)	3/23 (13%)
First incidence (days)	629	548	456	656
Life table test	P=0.043N	P=0.005N	P=0.007N	P=0.008N
Logistic regression test	P=0.037N	P=0.007N	P=0.031N	P=0.005N
Cochran-Armitage test	P=0.038N			
Fisher exact test		P=0.007N	P=0.132N	P=0.006N
Liver: Hepatocellular Adenoma				
Overall rate	0/50 (0%)	2/51 (4%)	5/50 (10%)	4/51 (8%)
Adjusted rate	0.0%	6.4%	13.5%	15.7%
Terminal rate	0/22 (0%)	1/26 (4%)	5/37 (14%)	3/23 (13%)
First incidence (days)	—[e]	645	740 (T)	720
Life table test	P=0.082	P=0.252	P=0.095	P=0.072
Logistic regression test	P=0.100	P=0.241	P=0.095	P=0.068
Cochran-Armitage test	P=0.104			
Fisher exact test		P=0.252	P=0.028	P=0.061

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Liver: Hepatocellular Adenoma or Carcinoma				
Overall rate	0/50 (0%)	2/51 (4%)	5/50 (10%)	5/51 (10%)
Adjusted rate	0.0%	6.4%	13.5%	18.5%
Terminal rate	0/22 (0%)	1/26 (4%)	5/37 (14%)	3/23 (13%)
First incidence (days)	—	645	740 (T)	720
Life table test	P=0.033	P=0.252	P=0.095	P=0.041
Logistic regression test	P=0.042	P=0.241	P=0.095	P=0.036
Cochran-Armitage test	P=0.045			
Fisher exact test		P=0.252	P=0.028	P=0.030
Lung: Alveolar/bronchiolar Adenoma or Carcinoma				
Overall rate	1/50 (2%)	1/51 (2%)	2/50 (4%)	3/51 (6%)
Adjusted rate	4.5%	3.8%	5.4%	10.0%
Terminal rate	1/22 (5%)	1/26 (4%)	2/37 (5%)	1/23 (4%)
First incidence (days)	740 (T)	740 (T)	740 (T)	590
Life table test	P=0.155	P=0.725N	P=0.678	P=0.314
Logistic regression test	P=0.176	P=0.725N	P=0.678	P=0.307
Cochran-Armitage test	P=0.183			
Fisher exact test		P=0.748N	P=0.500	P=0.316
Mammary Gland: Fibroadenoma				
Overall rate	21/50 (42%)	22/51 (43%)	23/50 (46%)	26/51 (51%)
Adjusted rate	62.0%	55.8%	51.9%	69.5%
Terminal rate	10/22 (45%)	10/26 (38%)	16/37 (43%)	12/23 (52%)
First incidence (days)	589	548	456	568
Life table test	P=0.189	P=0.510N	P=0.162N	P=0.292
Logistic regression test	P=0.161	P=0.516	P=0.526	P=0.207
Cochran-Armitage test	P=0.193			
Fisher exact test		P=0.534	P=0.420	P=0.240
Mammary Gland: Fibroadenoma or Adenoma				
Overall rate	22/50 (44%)	22/51 (43%)	23/50 (46%)	26/51 (51%)
Adjusted rate	63.3%	55.8%	51.9%	69.5%
Terminal rate	10/22 (45%)	10/26 (38%)	16/37 (43%)	12/23 (52%)
First incidence (days)	589	548	456	568
Life table test	P=0.224	P=0.444N	P=0.121N	P=0.356
Logistic regression test	P=0.196	P=0.565N	P=0.554N	P=0.272
Cochran-Armitage test	P=0.231			
Fisher exact test		P=0.545N	P=0.500	P=0.308
Mammary Gland: Carcinoma				
Overall rate	4/50 (8%)	5/51 (10%)	3/50 (6%)	3/51 (6%)
Adjusted rate	10.6%	15.7%	7.8%	8.1%
Terminal rate	0/22 (0%)	3/26 (12%)	2/37 (5%)	0/23 (0%)
First incidence (days)	481	628	722	506
Life table test	P=0.356N	P=0.530	P=0.359N	P=0.505N
Logistic regression test	P=0.316N	P=0.536	P=0.568N	P=0.407N
Cochran-Armitage test	P=0.346N			
Fisher exact test		P=0.513	P=0.500N	P=0.489N

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Mammary Gland: Adenoma or Carcinoma				
Overall rate	6/50 (12%)	5/51 (10%)	3/50 (6%)	3/51 (6%)
Adjusted rate	16.1%	15.7%	7.8%	8.1%
Terminal rate	0/22 (0%)	3/26 (12%)	2/37 (5%)	0/23 (0%)
First incidence (days)	481	628	722	506
Life table test	P=0.204N	P=0.479N	P=0.142N	P=0.248N
Logistic regression test	P=0.181N	P=0.469N	P=0.290N	P=0.184N
Cochran-Armitage test	P=0.199N			
Fisher exact test		P=0.486N	P=0.243N	P=0.234N
Mammary Gland: Fibroadenoma, Adenoma, or Carcinoma				
Overall rate	25/50 (50%)	27/51 (53%)	25/50 (50%)	28/51 (55%)
Adjusted rate	65.9%	65.7%	55.4%	70.9%
Terminal rate	10/22 (45%)	13/26 (50%)	17/37 (46%)	12/23 (52%)
First incidence (days)	481	548	456	506
Life table test	P=0.354	P=0.557N	P=0.087N	P=0.424
Logistic regression test	P=0.338	P=0.447	P=0.534N	P=0.337
Cochran-Armitage test	P=0.377			
Fisher exact test		P=0.462	P=0.579N	P=0.384
Pituitary Gland (Pars Distalis): Adenoma				
Overall rate	24/50 (48%)	23/51 (45%)	18/50 (36%)	20/51 (39%)
Adjusted rate	62.3%	64.9%	42.3%	55.0%
Terminal rate	9/22 (41%)	14/26 (54%)	13/37 (35%)	8/23 (35%)
First incidence (days)	537	601	610	448
Life table test	P=0.287N	P=0.374N	P=0.015N	P=0.275N
Logistic regression test	P=0.255N	P=0.484N	P=0.146N	P=0.269N
Cochran-Armitage test	P=0.232N			
Fisher exact test		P=0.463N	P=0.156N	P=0.245N
Pituitary Gland (Pars Distalis): Adenoma or Carcinoma				
Overall rate	24/50 (48%)	23/51 (45%)	18/50 (36%)	22/51 (43%)
Adjusted rate	62.3%	64.9%	42.3%	59.4%
Terminal rate	9/22 (41%)	14/26 (54%)	13/37 (35%)	9/23 (39%)
First incidence (days)	537	601	610	448
Life table test	P=0.441N	P=0.374N	P=0.015N	P=0.389N
Logistic regression test	P=0.429N	P=0.484N	P=0.146N	P=0.419N
Cochran-Armitage test	P=0.395N			
Fisher exact test		P=0.463N	P=0.156N	P=0.386N
Stomach (Forestomach): Squamous Cell Papilloma or Squamous Cell Carcinoma				
Overall rate	0/50 (0%)	3/51 (6%)	0/50 (0%)	2/51 (4%)
Adjusted rate	0.0%	11.5%	0.0%	6.5%
Terminal rate	0/22 (0%)	3/26 (12%)	0/37 (0%)	1/23 (4%)
First incidence (days)	—	740 (T)	—	590
Life table test	P=0.403	P=0.150	—	P=0.247
Logistic regression test	P=0.420	P=0.150	—	P=0.249
Cochran-Armitage test	P=0.426			
Fisher exact test		P=0.125	—	P=0.252

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Thyroid Gland (C-cell): Adenoma				
Overall rate	2/50 (4%)	2/51 (4%)	4/50 (8%)	6/51 (12%)
Adjusted rate	9.1%	7.7%	10.5%	20.2%
Terminal rate	2/22 (9%)	2/26 (8%)	3/37 (8%)	3/23 (13%)
First incidence (days)	740 (T)	740 (T)	722	656
Life table test	P=0.050	P=0.635N	P=0.572	P=0.159
Logistic regression test	P=0.059	P=0.635N	P=0.475	P=0.140
Cochran-Armitage test	P=0.062			
Fisher exact test		P=0.684N	P=0.339	P=0.141
Thyroid Gland (C-cell): Adenoma or Carcinoma				
Overall rate	2/50 (4%)	3/51 (6%)	5/50 (10%)	6/51 (12%)
Adjusted rate	9.1%	11.5%	12.5%	20.2%
Terminal rate	2/22 (9%)	3/26 (12%)	3/37 (8%)	3/23 (13%)
First incidence (days)	740 (T)	740 (T)	638	656
Life table test	P=0.091	P=0.577	P=0.418	P=0.159
Logistic regression test	P=0.100	P=0.577	P=0.264	P=0.140
Cochran-Armitage test	P=0.105			
Fisher exact test		P=0.509	P=0.218	P=0.141
Thyroid Gland (Follicular Cell): Adenoma or Carcinoma				
Overall rate	0/50 (0%)	1/51 (2%)	1/50 (2%)	3/51 (6%)
Adjusted rate	0.0%	3.8%	2.7%	10.1%
Terminal rate	0/22 (0%)	1/26 (4%)	1/37 (3%)	1/23 (4%)
First incidence (days)	—	740 (T)	740 (T)	720
Life table test	P=0.059	P=0.533	P=0.604	P=0.143
Logistic regression test	P=0.063	P=0.533	P=0.604	P=0.126
Cochran-Armitage test	P=0.066			
Fisher exact test		P=0.505	P=0.500	P=0.125
Uterus: Stromal Polyp				
Overall rate	12/50 (24%)	11/51 (22%)	7/50 (14%)	6/51 (12%)
Adjusted rate	33.3%	31.3%	17.4%	18.9%
Terminal rate	2/22 (9%)	4/26 (15%)	5/37 (14%)	1/23 (4%)
First incidence (days)	570	548	638	648
Life table test	P=0.078N	P=0.454N	P=0.050N	P=0.110N
Logistic regression test	P=0.067N	P=0.477N	P=0.170N	P=0.093N
Cochran-Armitage test	P=0.065N			
Fisher exact test		P=0.478N	P=0.154N	P=0.089N
All Organs: Mononuclear Cell Leukemia				
Overall rate	16/50 (32%)	21/51 (41%)	19/50 (38%)	16/51 (31%)
Adjusted rate	47.3%	49.8%	41.9%	40.8%
Terminal rate	6/22 (27%)	7/26 (27%)	11/37 (30%)	4/23 (17%)
First incidence (days)	589	308	532	441
Life table test	P=0.365N	P=0.308	P=0.345N	P=0.533N
Logistic regression test	P=0.311N	P=0.234	P=0.353	P=0.564N
Cochran-Armitage test	P=0.331N			
Fisher exact test		P=0.227	P=0.338	P=0.558N

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
All Organs: Benign Neoplasms				
Overall rate	42/50 (84%)	44/51 (86%)	41/50 (82%)	41/51 (80%)
Adjusted rate	93.2%	93.5%	87.2%	91.1%
Terminal rate	19/22 (86%)	23/26 (88%)	31/37 (84%)	19/23 (83%)
First incidence (days)	537	548	456	448
Life table test	P=0.447N	P=0.462N	P=0.006N	P=0.428N
Logistic regression test	P=0.424N	P=0.420	P=0.372N	P=0.565N
Cochran-Armitage test	P=0.294N			
Fisher exact test		P=0.483	P=0.500N	P=0.416N
All Organs: Malignant Neoplasms				
Overall rate	30/50 (60%)	29/51 (57%)	30/50 (60%)	28/51 (55%)
Adjusted rate	71.5%	64.1%	63.6%	64.4%
Terminal rate	11/22 (50%)	11/26 (42%)	20/37 (54%)	9/23 (39%)
First incidence (days)	481	308	456	441
Life table test	P=0.439N	P=0.399N	P=0.076N	P=0.398N
Logistic regression test	P=0.429N	P=0.432N	P=0.510	P=0.364N
Cochran-Armitage test	P=0.370N			
Fisher exact test		P=0.453N	P=0.581N	P=0.376N
All Organs: Benign or Malignant Neoplasms				
Overall rate	48/50 (96%)	50/51 (98%)	50/50 (100%)	49/51 (96%)
Adjusted rate	96.0%	98.0%	100.0%	98.0%
Terminal rate	20/22 (91%)	25/26 (96%)	37/37 (100%)	22/23 (96%)
First incidence (days)	481	308	456	441
Life table test	P=0.477	P=0.452N	P=0.011N	P=0.536N
Logistic regression test	P=0.607N	P=0.441	P=0.133	P=0.593
Cochran-Armitage test	P=0.518N			
Fisher exact test		P=0.492	P=0.247	P=0.684

(T)Terminal sacrifice
[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal gland, clitoral gland, liver, lung, pituitary gland, thyroid gland, and uterus; for other tissues, denominator is number of animals necropsied.
[b] Kaplan-Meier estimated neoplasm incidence at the end of the study after adjustment for intercurrent mortality
[c] Observed incidence at terminal kill
[d] Beneath the control incidence are the P values associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the controls and that exposed group. The life table test regards neoplasms in animals dying prior to terminal kill as being (directly or indirectly) the cause of death. The logistic regression test regards these lesions as nonfatal. The Cochran-Armitage and Fisher exact tests compare directly the overall incidence rates. For all tests, a negative trend or a lower incidence in an exposure group is indicated by **N**.
[e] Not applicable; no neoplasms in animal group

TABLE B4a
Historical Incidence of Hepatocellular Neoplasms in Untreated Female F344/N Rats [a]

Study	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	0/50	0/50	0/50
Benzyl Acetate	1/50	0/50	1/50
Butyl Benzyl Phthalate	0/50	0/50	0/50
C.I. Pigment Red 23	1/50	0/50	1/50
C.I. Pigment Red 3	0/50	0/50	0/50
o-Nitroanisole	0/50	0/50	0/50
p-Nitrobenzoic Acid	2/50	0/50	2/50
Polysorbate 80	0/50	0/50	0/50
Overall Historical Incidence			
Total	8/1,301 (0.6%)	1/1,301 (0.1%)	9/1,301 (0.7%)
Standard deviation	1.5%	0.4%	1.5%
Range	0%-6%	0%-2%	0%-6%

[a] Data as of 12 May 1995

TABLE B4b
Historical Incidence of Renal Tubule Neoplasms in Untreated Female F344/N Rats [a]

Study	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	0/50	0/50	0/50
Benzyl Acetate	0/50	0/50	0/50
Butyl Benzyl Phthalate	0/50	1/50	1/50
C.I. Pigment Red 23	0/50	0/50	0/50
C.I. Pigment Red 3	0/50	0/50	0/50
o-Nitroanisole	0/50	0/50	0/50
p-Nitrobenzoic Acid	0/50	0/50	0/50
Polysorbate 80	0/50	0/50	0/50
Overall Historical Incidence			
Total	0/1,298 (0%)	1/1,298 (0.1%)	1/1,298 (0.1%)
Standard deviation		0.4%	0.4%
Range		0%-2%	0%-2%

[a] Data as of 12 May 1995

TABLE B4c
Historical Incidence of Oral Cavity Neoplasms in Untreated Female F344/N Rats [a]

Study	Incidence in Controls		
	Squamous Cell Papilloma[b]	Squamous Cell Carcinoma	Squamous Cell Papilloma or Carcinoma[b]
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	2/50	0/50	2/50
Benzyl Acetate	1/50	0/50	1/50
Butyl Benzyl Phthalate	2/50	0/50	2/50
C.I. Pigment Red 23	0/50	0/50	0/50
C.I. Pigment Red 3	0/50	0/50	0/50
o-Nitroanisole	1/50	0/50	1/50
p-Nitrobenzoic Acid	0/50	0/50	0/50
Polysorbate 80	0/50	0/50	0/50
Overall Historical Incidence			
Total	11/1,301 (0.9%)	4/1,301 (0.3%)	15/1,301 (1.2%)
Standard Deviation	1.4%	0.7%	1.6%
Range	0%-4%	0%-2%	0%-6%

[a] Data as of 12 May 1995. Includes data for oral mucosa, tongue, pharynx, and tooth.
[b] Includes data for papilloma.

TABLE B4d
Historical Incidence of Clitoral Gland Neoplasms in Untreated Female F344/N Rats [a]

Study	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence at Southern Research Institute			
2,2-Bis(bromomethyl)-1,3-propanediol (FR-1138®)	4/48	1/48	5/48
Benzyl Acetate	0/50	1/50	1/50
Butyl Benzyl Phthalate	3/50	4/50	7/50
C.I. Pigment Red 23	5/47	3/47	7/47
C.I. Pigment Red 3	9/47	0/47	9/47
o-Nitroanisole	3/45	4/45	7/45
p-Nitrobenzoic Acid	4/50	1/50	4/50
Polysorbate 80	3/48	7/48	10/48
Overall Historical Incidence			
Total	99/1,218 (8.1%)	33/1,218 (2.7%)	130/1,218 (10.7%)
Standard deviation	4.1%	3.8%	5.3%
Range	0%-19%	0%-15%	2%-21%

[a] Data as of 12 May 1995

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Disposition Summary				
Animals initially in study	60	60	60	60
12-Month interim evaluation	10	9	10	9
Early deaths				
Moribund	25	23	12	25
Natural deaths	3	2	1	3
Survivors				
Terminal sacrifice	22	26	37	23
Animals examined microscopically	60	60	60	60
12-Month Interim Evaluation				
Alimentary System				
Intestine large, colon	(10)	(8)	(9)	(9)
Parasite metazoan	1 (10%)			1 (11%)
Intestine large, rectum	(10)	(9)	(10)	(9)
Parasite metazoan	2 (20%)			1 (11%)
Liver	(10)	(9)	(10)	(9)
Basophilic focus	7 (70%)	2 (22%)	2 (20%)	1 (11%)
Clear cell focus		1 (11%)	4 (40%)	4 (44%)
Eosinophilic focus			1 (10%)	
Granuloma			1 (10%)	
Hepatodiaphragmatic nodule		1 (11%)	2 (20%)	
Inflammation, subacute	3 (30%)	4 (44%)	7 (70%)	9 (100%)
Bile duct, hyperplasia	1 (10%)	1 (11%)	6 (60%)	9 (100%)
Bile duct, pigmentation		9 (100%)	7 (70%)	9 (100%)
Hepatocyte, cytologic alterations		4 (44%)	10 (100%)	9 (100%)
Hepatocyte, pigmentation		9 (100%)	10 (100%)	9 (100%)
Kupffer cell, pigmentation				9 (100%)
Mesentery	(1)	(2)		(2)
Accessory spleen				2 (100%)
Fat, necrosis	1 (100%)	2 (100%)		
Pancreas	(10)	(9)	(10)	(9)
Atrophy	2 (20%)		3 (30%)	1 (11%)
Salivary glands	(10)	(9)	(10)	(9)
Atrophy			1 (10%)	2 (22%)
Stomach, forestomach	(10)	(9)	(10)	(9)
Mucosa, hyperplasia			1 (10%)	
Stomach, glandular	(10)	(9)	(10)	(9)
Mineralization				2 (22%)
Cardiovascular System				
Heart	(10)	(9)	(10)	(9)
Cardiomyopathy	1 (10%)			
Endocrine System				
Adrenal cortex	(10)	(9)	(10)	(9)
Accessory adrenal cortical nodule		4 (44%)	2 (20%)	1 (11%)
Hypertrophy, focal			1 (10%)	

[a] Number of animals examined microscopically at the site and the number of animals with lesion

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
12-Month Interim Evaluation (continued)				
Endocrine System (continued)				
Pituitary gland	(10)	(9)	(10)	(9)
Pars distalis, angiectasis	1 (10%)		1 (10%)	1 (11%)
Pars distalis, cyst	4 (40%)	7 (78%)	9 (90%)	3 (33%)
Pars distalis, hyperplasia, focal	1 (10%)	2 (22%)	2 (20%)	1 (11%)
Pars intermedia, cyst	1 (10%)			
Pars intermedia, hyperplasia	1 (10%)			
Thyroid gland	(10)	(9)	(10)	(9)
Ultimobranchial cyst		2 (22%)	1 (10%)	1 (11%)
Genital System				
Clitoral gland	(10)	(9)	(10)	(9)
Hyperplasia			1 (10%)	
Inflammation, chronic				1 (11%)
Inflammation, chronic active		1 (11%)		
Ovary	(10)	(9)	(10)	(9)
Cyst	1 (10%)	3 (33%)	6 (60%)	1 (11%)
Uterus	(10)	(9)	(10)	(9)
Hydrometra		1 (11%)	1 (10%)	3 (33%)
Hematopoietic System				
Lymph node	(1)	(2)	(2)	(2)
Mediastinal, hemorrhage	1 (100%)	2 (100%)	2 (100%)	2 (100%)
Mediastinal, pigmentation	1 (100%)	2 (100%)	2 (100%)	2 (100%)
Lymph node, mandibular	(10)	(9)	(10)	(9)
Hemorrhage	3 (30%)		1 (10%)	
Pigmentation	1 (10%)	4 (44%)	1 (10%)	2 (22%)
Spleen	(10)	(9)	(10)	(9)
Hematopoietic cell proliferation	1 (10%)			1 (11%)
Pigmentation	7 (70%)	7 (78%)	6 (60%)	6 (67%)
Integumentary System				
Mammary gland	(10)	(9)	(10)	(9)
Hyperplasia	1 (10%)		1 (10%)	
Respiratory System				
Lung	(10)	(9)	(10)	(9)
Hemorrhage	1 (10%)			
Infiltration cellular, histiocyte	4 (40%)	1 (11%)	1 (10%)	2 (22%)
Inflammation, subacute		3 (33%)	1 (10%)	2 (22%)
Alveolar epithelium, hyperplasia	1 (10%)			
Special Senses System				
Eye			(1)	
Cataract			1 (100%)	
Hemorrhage			1 (100%)	
Retina, degeneration			1 (100%)	

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
12-Month Interim Evaluation (continued)				
Urinary System				
Kidney	(10)	(9)	(10)	(9)
Mineralization	10 (100%)	9 (100%)	7 (70%)	9 (100%)
Nephropathy	6 (60%)	6 (67%)	7 (70%)	9 (100%)
Renal tubule, pigmentation		9 (100%)	10 (100%)	9 (100%)
Transitional epithelium, hyperplasia				2 (22%)
Systems Examined With No Lesions Observed				
General Body System				
Musculoskeletal System				
Nervous System				
2-Year Study				
Alimentary System				
Intestine large, colon	(50)	(50)	(50)	(51)
Parasite metazoan	4 (8%)		4 (8%)	2 (4%)
Intestine large, rectum	(49)	(50)	(49)	(50)
Edema				1 (2%)
Parasite metazoan	3 (6%)	4 (8%)	6 (12%)	2 (4%)
Intestine large, cecum	(50)	(51)	(49)	(51)
Edema	1 (2%)	1 (2%)	2 (4%)	
Parasite metazoan	1 (2%)			
Ulcer				1 (2%)
Intestine small, duodenum	(50)	(51)	(50)	(50)
Ulcer		1 (2%)		
Epithelium, hyperplasia		1 (2%)		
Intestine small, jejunum	(49)	(51)	(50)	(51)
Epithelium, hyperplasia		1 (2%)		
Intestine small, ileum	(48)	(51)	(49)	(49)
Epithelium, hyperplasia		1 (2%)		
Liver	(50)	(51)	(50)	(51)
Angiectasis	1 (2%)	2 (4%)	3 (6%)	1 (2%)
Basophilic focus	32 (64%)	26 (51%)	11 (22%)	12 (24%)
Clear cell focus	10 (20%)	18 (35%)	29 (58%)	30 (59%)
Cyst		3 (6%)	4 (8%)	
Cytoplasmic alteration	2 (4%)			
Degeneration, cystic				1 (2%)
Eosinophilic focus	10 (20%)	9 (18%)	14 (28%)	16 (31%)
Granuloma	10 (20%)	1 (2%)	1 (2%)	1 (2%)
Hematopoietic cell proliferation	2 (4%)	1 (2%)	2 (4%)	7 (14%)
Hepatodiaphragmatic nodule	7 (14%)	4 (8%)	5 (10%)	5 (10%)
Inflammation, subacute	2 (4%)	1 (2%)	3 (6%)	3 (6%)
Mixed cell focus	12 (24%)	19 (37%)	20 (40%)	16 (31%)
Necrosis, focal	2 (4%)	2 (4%)	5 (10%)	4 (8%)
Thrombosis		1 (2%)	2 (4%)	2 (4%)
Bile duct, hyperplasia	14 (28%)	10 (20%)	27 (54%)	33 (65%)
Bile duct, pigmentation		46 (90%)	49 (98%)	50 (98%)
Centrilobular, atrophy	4 (8%)	9 (18%)	5 (10%)	5 (10%)
Centrilobular, necrosis	1 (2%)			1 (2%)
Hepatocyte, cytologic alterations		11 (22%)	31 (62%)	40 (78%)
Hepatocyte, pigmentation		34 (67%)	44 (88%)	50 (98%)
Hepatocyte, vacuolization cytoplasmic	6 (12%)	6 (12%)	2 (4%)	4 (8%)
Kupffer cell, pigmentation	9 (18%)	11 (22%)	16 (32%)	32 (63%)

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Alimentary System (continued)				
Mesentery	(11)	(18)	(8)	(12)
Accessory spleen	1 (9%)			
Angiectasis	1 (9%)			
Fat, necrosis	8 (73%)	16 (89%)	8 (100%)	11 (92%)
Pancreas	(50)	(51)	(50)	(51)
Atrophy	9 (18%)	19 (37%)	17 (34%)	14 (27%)
Necrosis			1 (2%)	
Acinar cell, cytoplasmic alteration	3 (6%)	2 (4%)		2 (4%)
Acinar cell, hyperplasia, focal	1 (2%)	1 (2%)		1 (2%)
Salivary glands	(50)	(46)	(50)	(50)
Atrophy		8 (17%)	7 (14%)	13 (26%)
Stomach, forestomach	(50)	(51)	(50)	(51)
Edema	5 (10%)		2 (4%)	4 (8%)
Fibrosis	1 (2%)			
Ulcer	4 (8%)	1 (2%)		3 (6%)
Mucosa, hyperplasia	5 (10%)	17 (33%)	30 (60%)	27 (53%)
Stomach, glandular	(50)	(51)	(50)	(51)
Edema		2 (4%)	2 (4%)	2 (4%)
Erosion	2 (4%)	1 (2%)		2 (4%)
Ulcer	1 (2%)	2 (4%)		
Tooth	(1)			
Developmental malformation	1 (100%)			
Cardiovascular System				
Blood vessel	(50)	(51)	(50)	(51)
Hypertrophy			1 (2%)	
Heart	(50)	(51)	(50)	(51)
Cardiomyopathy	16 (32%)	18 (35%)	18 (36%)	12 (24%)
Mineralization				1 (2%)
Thrombosis			1 (2%)	
Endocrine System				
Adrenal cortex	(50)	(51)	(50)	(51)
Accessory adrenal cortical nodule	5 (10%)	13 (25%)	11 (22%)	13 (25%)
Angiectasis		2 (4%)	2 (4%)	
Degeneration, fatty	9 (18%)	10 (20%)	9 (18%)	5 (10%)
Hematopoietic cell proliferation	1 (2%)	1 (2%)		1 (2%)
Hyperplasia, diffuse			1 (2%)	1 (2%)
Hyperplasia, focal	3 (6%)	2 (4%)	2 (4%)	1 (2%)
Hypertrophy	2 (4%)			
Hypertrophy, focal	3 (6%)	2 (4%)	2 (4%)	
Necrosis		1 (2%)		1 (2%)
Adrenal medulla	(48)	(51)	(50)	(51)
Hyperplasia	3 (6%)	1 (2%)	4 (8%)	
Islets, pancreatic	(50)	(51)	(49)	(51)
Hyperplasia	1 (2%)		1 (2%)	
Parathyroid gland	(48)	(48)	(48)	(50)
Hyperplasia		1 (2%)		

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Endocrine System (continued)				
Pituitary gland	(50)	(51)	(50)	(51)
Pars distalis, angiectasis	10 (20%)	6 (12%)	16 (32%)	12 (24%)
Pars distalis, cyst	25 (50%)	21 (41%)	25 (50%)	23 (45%)
Pars distalis, hyperplasia, focal	9 (18%)	4 (8%)	8 (16%)	8 (16%)
Pars intermedia, angiectasis	3 (6%)	1 (2%)	1 (2%)	
Pars intermedia, cyst			2 (4%)	2 (4%)
Pars intermedia, hyperplasia	1 (2%)			
Thyroid gland	(50)	(51)	(50)	(51)
Ultimobranchial cyst	3 (6%)	5 (10%)	4 (8%)	4 (8%)
C-cell, hyperplasia	13 (26%)	7 (14%)	10 (20%)	9 (18%)
Follicle, cyst			4 (8%)	4 (8%)
Follicular cell, hyperplasia	1 (2%)			
General Body System				
None				
Genital System				
Clitoral gland	(49)	(50)	(49)	(51)
Cyst	2 (4%)	4 (8%)	3 (6%)	1 (2%)
Hyperplasia	3 (6%)	1 (2%)	1 (2%)	1 (2%)
Inflammation, chronic				1 (2%)
Inflammation, suppurative	2 (4%)	1 (2%)		
Ovary	(50)	(51)	(50)	(51)
Cyst	13 (26%)	14 (27%)	11 (22%)	18 (35%)
Uterus	(50)	(51)	(50)	(51)
Hydrometra	2 (4%)	8 (16%)	3 (6%)	7 (14%)
Hyperplasia, cystic	5 (10%)	1 (2%)	4 (8%)	
Hematopoietic System				
Bone marrow	(50)	(51)	(50)	(51)
Hyperplasia	4 (8%)	1 (2%)	3 (6%)	5 (10%)
Infiltration cellular, histiocyte	1 (2%)			
Myelofibrosis	3 (6%)	2 (4%)	3 (6%)	3 (6%)
Necrosis	1 (2%)			
Lymph node	(9)	(11)	(11)	(15)
Deep cervical, pigmentation		1 (9%)		
Iliac, pigmentation				1 (7%)
Inguinal, hyperplasia, lymphoid	1 (11%)			
Mediastinal, hemorrhage	1 (11%)	2 (18%)	3 (27%)	2 (13%)
Mediastinal, hyperplasia, lymphoid		2 (18%)	1 (9%)	1 (7%)
Mediastinal, pigmentation	5 (56%)	7 (64%)	6 (55%)	8 (53%)
Pancreatic, granuloma				1 (7%)
Pancreatic, hemorrhage	1 (11%)			2 (13%)
Pancreatic, hyperplasia, lymphoid	1 (11%)		1 (9%)	2 (13%)
Pancreatic, pigmentation	1 (11%)	2 (18%)	6 (55%)	8 (53%)
Renal, hemorrhage		1 (9%)		1 (7%)
Renal, hyperplasia, lymphoid				1 (7%)
Renal, pigmentation		1 (9%)		1 (7%)

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Hematopoietic System (continued)				
Lymph node, mandibular	(50)	(51)	(50)	(49)
Ectasia	1 (2%)	3 (6%)	7 (14%)	5 (10%)
Hemorrhage	10 (20%)	11 (22%)	16 (32%)	7 (14%)
Hyperplasia, lymphoid	5 (10%)	10 (20%)	14 (28%)	13 (27%)
Pigmentation	23 (46%)	24 (47%)	20 (40%)	19 (39%)
Lymph node, mesenteric	(50)	(51)	(50)	(51)
Hemorrhage	5 (10%)	1 (2%)	1 (2%)	2 (4%)
Hyperplasia, lymphoid	2 (4%)	5 (10%)	5 (10%)	2 (4%)
Pigmentation		1 (2%)		1 (2%)
Spleen	(50)	(50)	(50)	(51)
Congestion				1 (2%)
Developmental malformation				1 (2%)
Fibrosis		3 (6%)	5 (10%)	4 (8%)
Hematopoietic cell proliferation	27 (54%)	23 (46%)	30 (60%)	31 (61%)
Infiltration cellular, histiocyte	1 (2%)			
Metaplasia, osseous		1 (2%)		
Necrosis	1 (2%)		1 (2%)	1 (2%)
Pigmentation	30 (60%)	30 (60%)	26 (52%)	32 (63%)
Lymphoid follicle, atrophy			2 (4%)	
Thymus	(50)	(49)	(49)	(49)
Hemorrhage		1 (2%)		
Hyperplasia				1 (2%)
Integumentary System				
Mammary gland	(50)	(51)	(50)	(51)
Hyperplasia	46 (92%)	42 (82%)	36 (72%)	42 (82%)
Skin	(49)	(51)	(50)	(51)
Subcutaneous tissue, edema				1 (2%)
Musculoskeletal System				
Bone	(50)	(51)	(50)	(51)
Arthrosis				1 (2%)
Fibrous osteodystrophy	1 (2%)			
Hyperostosis		1 (2%)		
Cranium, osteopetrosis	6 (12%)	7 (14%)	3 (6%)	3 (6%)
Femur, osteopetrosis	5 (10%)	7 (14%)	1 (2%)	2 (4%)
Nervous System				
Brain	(50)	(51)	(50)	(51)
Developmental malformation	9 (18%)	11 (22%)	8 (16%)	14 (27%)
Gliosis				1 (2%)
Hemorrhage				1 (2%)
Hydrocephalus	2 (4%)			4 (8%)
Peripheral nerve	(2)	(2)		(2)
Degeneration		1 (50%)		
Inflammation, chronic		1 (50%)		
Spinal cord	(3)	(2)		(2)
Necrosis	1 (33%)			

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 (continued)

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
2-Year Study (continued)				
Respiratory System				
Lung	(50)	(51)	(50)	(51)
Edema				1 (2%)
Hemorrhage			1 (2%)	6 (12%)
Infiltration cellular, histiocyte	34 (68%)	33 (65%)	36 (72%)	41 (80%)
Inflammation, subacute		3 (6%)		2 (4%)
Alveolar epithelium, hyperplasia		2 (4%)	3 (6%)	6 (12%)
Nose	(50)	(51)	(50)	(51)
Exudate	5 (10%)	5 (10%)	3 (6%)	2 (4%)
Foreign body	2 (4%)	1 (2%)	2 (4%)	
Mucosa, hyperplasia	4 (8%)	2 (4%)	2 (4%)	
Mucosa, metaplasia, squamous	3 (6%)	2 (4%)	2 (4%)	1 (2%)
Special Senses System				
Eye	(1)	(2)		
Atrophy	1 (100%)			
Cataract		2 (100%)		
Retina, degeneration		2 (100%)		
Urinary System				
Kidney	(50)	(51)	(50)	(51)
Inflammation, suppurative		1 (2%)		
Mineralization	48 (96%)	49 (96%)	47 (94%)	33 (65%)
Nephropathy	45 (90%)	47 (92%)	46 (92%)	50 (98%)
Renal tubule, cytoplasmic alteration	1 (2%)	2 (4%)	1 (2%)	1 (2%)
Renal tubule, necrosis	1 (2%)			1 (2%)
Renal tubule, pigmentation	10 (20%)	48 (94%)	50 (100%)	51 (100%)
Transitional epithelium, hyperplasia	2 (4%)	6 (12%)	10 (20%)	3 (6%)
Urinary bladder	(50)	(51)	(50)	(51)
Hyperplasia				1 (2%)

APPENDIX C
GENETIC TOXICOLOGY

SALMONELLA MUTAGENICITY TEST PROTOCOL 140
CHINESE HAMSTER OVARY CELL CYTOGENETICS PROTOCOLS 140
MOUSE PERIPHERAL BLOOD MICRONUCLEUS TEST PROTOCOL 141
RESULTS 142
TABLE C1 Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* 143
TABLE C2 Induction of Sister Chromatid Exchanges in Chinese Hamster Ovary Cells by D&C Yellow No. 11 150
TABLE C3 Induction of Chromosomal Aberrations in Chinese Hamster Ovary Cells by D&C Yellow No. 11 151
TABLE C4 Frequency of Micronuclei in Mouse Peripheral Blood Erythrocytes Following Treatment with D&C Yellow No. 11 in Feed for 13 Weeks 152

GENETIC TOXICOLOGY

SALMONELLA MUTAGENICITY TEST PROTOCOL

Testing was performed as reported by Zeiger *et al.* (1988). D&C Yellow No. 11 was sent to the laboratories as a coded aliquot from Radian Corporation (Austin, TX). It was incubated with the *Salmonella typhimurium* tester strains TA97, TA98, TA100, TA1535, and TA1538 either in buffer or S9 mix (metabolic activation enzymes and cofactors from Aroclor 1254-induced male Sprague-Dawley rat or Syrian hamster liver) for 20 minutes at 37 ° C. Top agar supplemented with *l*-histidine and *d*-biotin was added, and the contents of the tubes were mixed and poured onto the surfaces of minimal glucose agar plates. Histidine-independent mutant colonies arising on these plates were counted following incubation for 2 days at 37 ° C.

Each trial consisted of triplicate plates of concurrent positive and negative controls and at least five doses of D&C Yellow No. 11. By study design, 10,000 µg/plate was selected as the high dose in the study conducted at SRI International, and 4,000 µg/plate was selected in the study conducted at Microbiological Associates, Inc. All positive trials were repeated under the conditions that elicited the positive response.

In this assay, a positive response is defined as a reproducible, dose-related increase in histidine-independent (revertant) colonies in any one strain/activation combination. An equivocal response is defined as an increase in revertants that is not dose related, is not reproducible, or is of insufficient magnitude to support a determination of mutagenicity. A negative response is obtained when no increase in revertant colonies is observed following chemical treatment. There is no minimum percentage or fold increase required for a chemical to be judged positive or weakly positive.

CHINESE HAMSTER OVARY CELL CYTOGENETICS PROTOCOLS

Testing was performed as reported by Galloway *et al.* (1987). D&C Yellow No. 11 was sent to the laboratory as a coded aliquot by Radian Corporation. It was tested in cultured Chinese hamster ovary (CHO) cells for induction of sister chromatid exchanges (SCEs) and chromosomal aberrations (Abs), both in the presence and absence of Aroclor 1254-induced male Sprague-Dawley rat liver S9 and cofactor mix. Cultures were handled under gold lights to prevent photolysis of bromodeoxyuridine-substituted DNA. Each test consisted of concurrent solvent and positive controls and of at least three doses of D&C Yellow No. 11; the high dose was limited by toxicity. A single flask per dose was used.

Sister Chromatid Exchange Test: In the SCE test without S9, CHO cells were incubated for 26 hours with D&C Yellow No. 11 in McCoy's 5A medium supplemented with fetal bovine serum, *l*-glutamine, and antibiotics. Bromodeoxyuridine (BrdU) was added 2 hours after culture initiation. After 26 hours, the medium containing D&C Yellow No. 11 was removed and replaced with fresh medium plus BrdU and Colcemid, and incubation was continued for 2 hours. Cells were then harvested by mitotic shake-off, fixed, and stained with Hoechst 33258 and Giemsa. In the SCE test with S9, cells were incubated with D&C Yellow No. 11, serum-free medium, and S9 for 2 hours. The medium was then removed and replaced with medium containing serum and BrdU and no D&C Yellow No. 11, and incubation proceeded for an additional 26 hours with Colcemid present for the final 2 hours. Harvesting and staining were the same as for cells treated without S9. All slides were scored blind, and those from a single test were read by the same person. Fifty second-division metaphase cells were scored for frequency of SCEs/cell from each dose level. Because significant chemical-induced cell cycle delay was seen at some dose levels, incubation time was lengthened for these cultures to ensure a sufficient number of scorable (second-division metaphase) cells.

Statistical analyses were conducted on the slopes of the dose-response curves and the individual dose points (Galloway *et al.*, 1987). An SCE frequency 20% above the concurrent solvent control value was chosen as a statistically conservative positive response. The probability of this level of difference occurring by chance at one dose point is less than 0.01; the probability for such a chance occurrence at two dose points is less than 0.001. An increase of 20% or greater at any single dose was considered weak evidence of activity; increases at two or more doses resulted in a determination that the trial was positive. A statistically significant trend ($P<0.005$) in the absence of any responses reaching 20% above background led to a call of equivocal.

Chromosomal Aberrations Test: In the Abs test without S9, cells were incubated in McCoy's 5A medium with D&C Yellow No. 11 for 8.5 hours; Colcemid was added, and incubation continued for 2 hours. The cells were then harvested by mitotic shake-off, fixed, and stained with Giemsa. For the Abs test with S9, cells were treated with D&C Yellow No. 11 and S9 for 2 hours, after which the treatment medium was removed and the cells were incubated for 19.5 hours in fresh medium with Colcemid present for the final 2 hours. Cells were harvested in the same manner as for the treatment without S9. The harvest time for the Abs test was based on the cell cycle information obtained in the SCE test; because cell cycle delay was anticipated, the incubation period was extended.

Cells were selected for scoring on the basis of good morphology and completeness of karyotype (21 ± 2 chromosomes). All slides were scored blind, and those from a single test were read by the same person. Two hundred first-division metaphase cells were scored in the low-dose control cultures and the lowest dose in the initial trial without S9. Because high numbers of aberrations were observed, making a smaller sample size necessary for statistical precision and making the scoring process difficult, fewer cells (25 to 100) were scored in the other cultures. Classes of aberrations included simple (breaks and terminal deletions), complex (rearrangements and translocations), and other (pulverized cells, despiralized chromosomes, and cells containing 10 or more aberrations).

Chromosomal aberration data are presented as percentages of cells with aberrations. To arrive at a statistical call for a trial, analyses were conducted on both the dose-response curve and individual dose points. For a single trial, a statistically significant ($P \leq 0.05$) difference for one dose point and a significant trend ($P \leq 0.015$) were considered weak evidence for a positive response; significant differences for two or more doses indicated the trial was positive. A positive trend test in the absence of a statistically significant increase at any one dose resulted in an equivocal call (Galloway *et al.*, 1987). Ultimately, the trial calls were based on a consideration of the statistical analyses as well as the biological information available to the reviewers.

MOUSE PERIPHERAL BLOOD MICRONUCLEUS TEST PROTOCOL

A detailed discussion of this assay is presented in MacGregor *et al.* (1990). At the end of a 13-week toxicity study (NTP, 1991a), peripheral blood samples were obtained from male and female $B6C3F_1$ mice, and smears were immediately prepared and fixed in absolute methanol. The methanol-fixed slides were stained with a chromatin-specific fluorescent dye mixture of Hoechst 33258/pyronin Y (MacGregor *et al*., 1983) and coded. Slides were scanned at 630 or 1,000× magnification using a semi-automated image analysis system to determine the frequency of micronuclei in 10,000 normochromatic erythrocytes (NCEs) in each of 9 or 10 animals per exposure group. The criteria of Schmid (1976) were used to define micronuclei, with the additional requirement that the micronuclei exhibit the characteristic fluorescent emissions of DNA (blue with 360 nm and orange with 540 nm ultraviolet illumination); the minimum size was approximately one-twentieth the diameter of the NCE.

Log transformation of the NCE data, testing of normality by the Shapiro-Wilk test, and testing for heterogeneity of variance by Cochran's test were performed before statistical analyses. The frequency of micronucleated cells among NCEs was analyzed by analysis of variance using the SAS GLM procedure. The NCE data for each exposure group were compared with the concurrent control using Student's *t*-test.

RESULTS

Results of mutagenicity tests with D&C Yellow No. 11 in *S. typhimurium* were equivocal in the SRI International study, based on the responses observed in strain TA100 with 10% induced rat liver S9, and the results were weakly positive in the Microbiological Associates, Inc., study, which used slightly lower doses, based on responses observed in strains TA98 and TA100 with 30% induced rat or hamster liver S9 (Table C1; Zeiger *et al.*, 1988). No indication of mutagenic activity was observed in the absence of S9 in any of the strains tested. The data from the *S. typhimurium* studies indicate variable responses among replicate trials within a particular treatment condition; this may have been the result of precipitate formation at higher concentrations (333 µg/plate and above) and consequent variability in the actual D&C Yellow No. 11 exposure concentrations.

In cytogenic tests with cultured CHO cells, D&C Yellow No. 11 induced highly significant increases in both SCEs (Table C2) and Abs (Table C3) with and without S9. Cell cycle delay, requiring an extended incubation period, was observed in the SCE test at doses of 1.5 µg/mL and above; in the Abs test, no delay was observed in the absence of S9, but cultures treated in the presence of S9 were harvested late because cell cycle delay was anticipated. Less than 200 cells per dose level were scored at all but one dose level in the Abs test due to the high number of Abs per cell (cultures treated with S9), the frequency of aberrant cells, and the difficulty in finding scorable cells in some cases (Trial 1, without S9).

Despite the strong response seen in the *in vitro* Abs assay, no increase in the frequency of micronucleated NCEs was observed in peripheral blood samples from male and female mice given D&C Yellow No. 11 in feed for 13 weeks (Table C4).

In conclusion, D&C Yellow No. 11 was mutagenic in bacteria and clastogenic in mammalian cells *in vitro*, but no evidence of clastogenicity was observed in the single *in vivo* study performed with male and female mice.

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium*[a]

Strain	Dose (µg/plate)	Revertants/plate[b] −S9 Trial 1	−S9 Trial 2	−S9 Trial 3	+hamster S9 10%	+hamster S9 10%	+hamster S9 30%
Study performed at SRI International							
TA100	0	120 ± 7.4	109 ± 5.8	109 ± 11.0	132 ± 14.1	115 ± 5.8	119 ± 7.4
	1		92 ± 10.7			114 ± 13.3	
	3		96 ± 13.1	94 ± 5.8		116 ± 4.2	133 ± 16.6
	6		87 ± 11.6			112 ± 10.2	
	10		104 ± 10.4	110 ± 5.9		128 ± 6.7	133 ± 9.9
	33		121 ± 8.2	138 ± 7.8		148 ± 5.0	157 ± 6.9
	100	118 ± 7.1		95 ± 6.7	169 ± 2.9		169 ± 10.3
	333	95 ± 5.2[c]		80 ± 5.9[c]	151 ± 10.9[c]		131 ± 6.8[c]
	1,000	91 ± 4.8[c]			136 ± 8.1[c]		
	3,333	93 ± 5.8[c]			158 ± 7.5[c]		
	10,000	111 ± 3.2[c]			169 ± 6.4[c]		
Trial summary		Negative	Negative	Negative	Negative	Negative	Equivocal
Positive control[d]		629 ± 23.5	438 ± 19.3	554 ± 21.5	683 ± 31.0	989 ± 51.7	622 ± 79.8

		+rat S9 5%	+rat S9 5%	+rat S9 10%	+rat S9 10%	+rat S9 10%	+rat S9 10%
TA100 (continued)	0	121 ± 6.5	91 ± 5.9	95 ± 7.8	147 ± 4.8	143 ± 10.4	144 ± 7.0
	0.3		108 ± 11.2				
	1		87 ± 8.1				
	3		101 ± 8.1		127 ± 13.0		
	10		121 ± 25.2		146 ± 4.5		181 ± 14.5
	16						208 ± 11.0
	20						211 ± 16.7
	33		142 ± 16.6		208 ± 21.2		184 ± 7.2
	66						182 ± 17.5
	100	167 ± 5.9		196 ± 10.8	164 ± 16.9	178 ± 14.3	186 ± 8.5
	166				142 ± 7.5[c]		182 ± 9.0[c]
	200						173 ± 7.0[c]
	333	141 ± 18.9[c]		164 ± 7.6[c]		182 ± 9.8[c]	165 ± 9.7[c]
	1,000	153 ± 6.1[c]		154 ± 10.6[c]		185 ± 6.8[c]	
	3,333	150 ± 10.9[c]		186 ± 8.8[c]		184 ± 7.5[c]	
	10,000	161 ± 6.2[c]		202 ± 8.7[c]		173 ± 7.1[c]	
Trial summary		Negative	Equivocal	Equivocal	Equivocal	Equivocal	Equivocal
Positive control		1,276 ± 45.0	889 ± 53.5	445 ± 9.2	470 ± 14.8	502 ± 24.0	776 ± 21.9

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* (continued)

Strain	Dose (µg/plate)	Revertants/plate +rat S9 (continued) 10%	10%	10%	30%	30%	30%
Study performed at SRI International (continued)							
TA100 (continued)	0	135 ± 2.7	113 ± 10.7	86 ± 5.0	141 ± 5.5	112 ± 6.1	112 ± 12.2
	0.3	116 ± 1.3		88 ± 6.8		97 ± 9.1	
	1	115 ± 7.7	115 ± 11.2	79 ± 3.8		90 ± 7.6	
	3	151 ± 5.5	121 ± 4.8	93 ± 4.3		84 ± 1.5	94 ± 4.4
	6		115 ± 5.9				
	10	200 ± 16.3	151 ± 11.3	112 ± 9.0		85 ± 4.3	107 ± 5.2
	33	213 ± 11.1	181 ± 5.0	142 ± 13.3		120 ± 19.7	123 ± 6.2
	100				159 ± 5.0		178 ± 10.6
	333				164 ± 2.5[c]		154 ± 14.0[c]
	1,000				174 ± 12.0[c]		
	3,333				149 ± 8.0[c]		
	10,000				169 ± 3.4[c]		
Trial summary		Equivocal	Equivocal	Equivocal	Negative	Negative	Equivocal
Positive control		114 ± 4.5	567 ± 5.6	421 ± 27.9	307 ± 20.8	296 ± 11.0	245 ± 12.2

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* (continued)

Strain	Dose (µg/plate)	Revertants/plate −S9 Trial 1	−S9 Trial 2	−S9 Trial 3	+hamster S9 10%	+hamster S9 10%	+hamster S9 30%
Study performed at SRI International (continued)							
TA1535	0	16 ± 1.5	16 ± 1.5	21 ± 0.3	7 ± 1.2	8 ± 2.0	10 ± 1.8
	1		17 ± 3.3			9 ± 2.1	
	3		17 ± 3.8	14 ± 2.7		13 ± 2.6	7 ± 0.6
	6		15 ± 0.0			9 ± 2.3	
	10		15 ± 1.5	15 ± 3.2		13 ± 0.3	6 ± 1.2
	33		20 ± 2.3	13 ± 2.3		10 ± 0.9	8 ± 1.8
	100	6 ± 0.9		11 ± 1.0	8 ± 1.2		7 ± 0.3
	333	8 ± 1.5[c]		9 ± 1.8[c]	6 ± 0.7[c]		5 ± 1.2[c]
	1,000	7 ± 0.6[c]			7 ± 2.0[c]		
	3,333	6 ± 0.3[c]			9 ± 0.3[c]		
	10,000	8 ± 3.2[c]			8 ± 2.0[c]		
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control		434 ± 11.3	417 ± 13.9	442 ± 7.5	333 ± 31.3	483 ± 10.7	432 ± 55.5

		+rat S9 10%	+rat S9 10%	+rat S9 30%
TA1535 (continued)	0	9 ± 1.5	10 ± 2.3	13 ± 2.0
	1		11 ± 1.8	
	3		11 ± 1.5	10 ± 2.0
	6		11 ± 1.9	
	10		9 ± 1.3	8 ± 1.8
	33		8 ± 0.6	10 ± 2.7
	100	11 ± 2.9		12 ± 1.5
	333	5 ± 0.6[c]		7 ± 0.7[c]
	1,000	5 ± 1.2[c]		
	3,333	9 ± 1.3[c]		
	10,000	10 ± 1.5[c]		
Trial summary		Negative	Negative	Negative
Positive control		158 ± 7.4	290 ± 5.5	97 ± 10.5

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* (continued)

Strain	Dose (µg/plate)	Revertants/plate −S9 Trial 1	−S9 Trial 2	−S9 Trial 3	+hamster S9 10%	+hamster S9 10%	+hamster S9 30%
Study performed at SRI International (continued)							
TA97	0	148 ± 4.7	129 ± 7.9	137 ± 10.9	189 ± 7.0	158 ± 6.1	159 ± 12.5
	1		115 ± 9.8			167 ± 4.7	
	3		118 ± 10.7	150 ± 8.6		168 ± 5.2	153 ± 3.2
	6		112 ± 7.2			166 ± 3.4	
	10		109 ± 16.7	131 ± 10.5		176 ± 6.1	182 ± 21.5
	33		112 ± 15.6	147 ± 10.8		174 ± 9.7	211 ± 4.0
	100	154 ± 4.3		98 ± 7.0	202 ± 9.1		181 ± 35.9
	333	129 ± 3.4[c]		66 ± 25.5[c]	169 ± 8.7[c]		109 ± 17.8[c]
	1,000	113 ± 2.6[c]			168 ± 17.9[c]		
	3,333	111 ± 14.2[c]			209 ± 3.8[c]		
	10,000	141 ± 13.6[c]			197 ± 5.7[c]		
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control		352 ± 22.5	579 ± 5.7	795 ± 35.9	1,060 ± 75.7	528 ± 26.9	536 ± 4.9

		+rat S9 10%	+rat S9 10%	+rat S9 30%
TA97 (continued)	0	200 ± 5.8	160 ± 2.9	210 ± 8.1
	1		159 ± 10.7	
	3		173 ± 5.5	184 ± 14.1
	6		169 ± 2.2	
	10		185 ± 12.8	175 ± 7.4
	33		217 ± 22.6	209 ± 5.8
	100	192 ± 3.4		224 ± 5.1
	333	171 ± 29.9[c]		114 ± 4.0[c]
	1,000	147 ± 14.7[c]		
	3,333	186 ± 9.8[c]		
	10,000	187 ± 25.0[c]		
Trial summary		Negative	Equivocal	Negative
Positive control		365 ± 17.8	375 ± 13.5	469 ± 11.3

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* (continued)

Strain	Dose (µg/plate)	Revertants/plate −S9 Trial 1	−S9 Trial 2	−S9 Trial 3	+hamster S9 10%	+hamster S9 10%	+hamster S9 30%
Study performed at SRI International (continued)							
TA98	0	17 ± 3.5	18 ± 0.7	24 ± 2.3	37 ± 4.1	31 ± 2.6	34 ± 0.0
	1		19 ± 1.2			27 ± 3.4	
	3		17 ± 1.3	19 ± 1.5		33 ± 2.1	30 ± 3.5
	6		20 ± 1.2			29 ± 2.4	
	10		15 ± 1.0	20 ± 2.4		31 ± 2.5	37 ± 4.4
	33		17 ± 1.8	15 ± 0.7		46 ± 4.7	42 ± 2.8
	100	17 ± 3.3		16 ± 1.9	45 ± 4.3		49 ± 7.5
	333	10 ± 1.2[c]		12 ± 1.5[c]	26 ± 2.6[c]		36 ± 2.3[c]
	1,000	11 ± 2.9[c]			29 ± 6.0[c]		
	3,333	12 ± 0.7[c]			54 ± 4.0[c]		
	10,000	19 ± 0.7[c]			52 ± 2.2[c]		
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control		751 ± 2.0	569 ± 20.3	595 ± 29.0	594 ± 51.4	782 ± 20.3	468 ± 30.7

Strain	Dose (µg/plate)	+rat S9 5%	+rat S9 10%	+rat S9 10%	+rat S9 10%	+rat S9 10%	+rat S9 30%	+rat S9 30%
TA98 (continued)	0	28 ± 5.3	28 ± 4.7	22 ± 3.1	30 ± 2.0	26 ± 1.9	29 ± 4.7	23 ± 5.0
	1					28 ± 3.8		
	3			27 ± 1.5		28 ± 5.1		27 ± 4.8
	6					29 ± 1.9		
	10			29 ± 2.7		30 ± 3.3		27 ± 6.1
	33			36 ± 7.1		43 ± 2.9		28 ± 4.6
	100	42 ± 1.7	56 ± 4.7	37 ± 2.1	37 ± 4.2		38 ± 4.1	38 ± 2.3
	166			28 ± 7.0[c]				
	333	41 ± 4.0[c]	26 ± 3.3[c]		34 ± 2.1[c]		32 ± 5.8[c]	26 ± 1.5[c]
	1,000	32 ± 3.7[c]	26 ± 5.8[c]		37 ± 3.9[c]		29 ± 6.7[c]	
	3,333	30 ± 1.8[c]	42 ± 4.6[c]		39 ± 1.7[c]		36 ± 4.1[c]	
	10,000	34 ± 3.8[c]	53 ± 2.4[c]		44 ± 4.4[c]		38 ± 4.9[c]	
Trial summary		Negative	Equivocal	Equivocal	Negative	Negative	Negative	Negative
Positive control		681 ± 33.5	335 ± 6.6	293 ± 6.1	394 ± 5.6	514 ± 3.0	62 ± 4.5	135 ± 12.1

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* (continued)

Strain	Dose (µg/plate)	Revertants/plate −S9	+hamster S9 30%	+hamster S9 30%
Study performed at Microbiological Associates, Inc.				
TA100	0	87 ± 4.2	108 ± 4.6	97 ± 2.3
	3.3			97 ± 4.7
	10	97 ± 3.0	106 ± 0.9	111 ± 3.5
	33	92 ± 7.5	141 ± 2.3	125 ± 6.4
	100	90 ± 5.3	151 ± 7.1	181 ± 7.1
	333	74 ± 3.5	154 ± 6.1	186 ± 11.0[c]
	1,000			200 ± 40.5[c]
	3,333	89 ± 10.2[c]	154 ± 3.5[c]	195 ± 11.0[c]
	4,000			190 ± 4.7[c]
Trial summary		Negative	Equivocal	Positive
Positive control		560 ± 10.1	531 ± 14.2	461 ± 45.5

Strain	Dose (µg/plate)	+rat S9 5%	+rat S9 10%	+rat S9 30%	+rat S9 30%	+rat S9 30%
TA100 (continued)	0	109 ± 6.0	110 ± 6.9	112 ± 8.1	116 ± 4.5	97 ± 3.2
	3.3					83 ± 1.7
	10			109 ± 10.0		92 ± 4.6
	33			123 ± 4.4		115 ± 13.6
	100	122 ± 6.4	129 ± 8.9	171 ± 5.7	184 ± 11.1	199 ± 1.8
	333	112 ± 4.0	119 ± 2.8	162 ± 10.1	171 ± 7.5	211 ± 13.4[c]
	1,000	117 ± 2.8	128 ± 9.9		159 ± 4.1	217 ± 20.0[c]
	3,333	113 ± 4.2	126 ± 4.2	179 ± 7.7[c]	168 ± 1.2	217 ± 11.6[c]
	4,000	120 ± 6.0	142 ± 12.6		155 ± 9.8	211 ± 2.9[c]
Trial summary		Negative	Negative	Weakly positive	Equivocal	Positive
Positive control		530 ± 57.3	280 ± 16.8	331 ± 0.0	411 ± 45.8	438 ± 15.8

Strain	Dose (µg/plate)	+ S9 30% hamster	+ S9 30% rat
TA1538	0	16 ± 2.3	16 ± 2.2
	3.3	15 ± 2.0	16 ± 3.8
	10	18 ± 2.1	16 ± 0.3
	33	24 ± 2.1	16 ± 5.2
	100	28 ± 0.9	28 ± 1.5
	333	30 ± 2.2[c]	32 ± 2.7[c]
	1,000	30 ± 4.2[c]	23 ± 3.1[c]
	3,333	37 ± 2.9[c]	24 ± 1.5[c]
	4,000	29 ± 3.5[c]	28 ± 2.2[c]
Trial summary		Weakly positive	Equivocal
Positive control		108 ± 8.0	97 ± 3.1

TABLE C1
Mutagenicity of D&C Yellow No. 11 in *Salmonella typhimurium* (continued)

Strain	Dose (µg/plate)	Revertants/plate −S9	+hamster S9 5%	+hamster S9 10%	+hamster S9 30%	+hamster S9 30%	+hamster S9 30%	+hamster S9 30%
Study performed at Microbiological Associates, Inc. (continued)								
TA98	0	15 ± 3.5	18 ± 0.5	24 ± 3.1	30 ± 1.9	23 ± 1.5	22 ± 2.0	29 ± 1.2
	3.3							31 ± 3.2
	10	15 ± 3.7			32 ± 4.0			30 ± 2.1
	33	14 ± 2.0			35 ± 1.2			34 ± 3.8
	100	13 ± 3.2	26 ± 3.6	44 ± 2.0	51 ± 5.9	65 ± 3.8	39 ± 3.5	59 ± 2.6
	333	14 ± 1.5	25 ± 2.3	39 ± 3.5	46 ± 3.2	52 ± 7.5	52 ± 4.9	49 ± 3.3[c]
	1,000		27 ± 4.8	40 ± 3.5		62 ± 1.7	53 ± 4.1[c]	64 ± 5.0[c]
	3,333	17 ± 2.7[c]	28 ± 1.8	44 ± 3.1	47 ± 2.3[c]	63 ± 2.0	52 ± 5.0[c]	66 ± 3.5[c]
	4,000		31 ± 2.0	43 ± 0.7		54 ± 3.6	52 ± 8.5[c]	65 ± 1.2[c]
Trial summary		Negative	Negative	Equivocal	Negative	Weakly positive	Positive	Positive
Positive control		254 ± 3.8	93 ± 24.2	101 ± 20.9	75 ± 1.2	56 ± 2.0	53 ± 5.2	98 ± 4.1

		+rat S9 5%	+rat S9 10%	+rat S9 30%	+rat S9 30%	+rat S9 30%	+rat S9 30%
TA98 (continued)	0	25 ± 5.7	30 ± 4.8	27 ± 3.4	21 ± 1.7	31 ± 3.0	27 ± 2.3
	3.3						23 ± 3.8
	10			22 ± 4.9			26 ± 2.7
	33			34 ± 2.5			28 ± 4.0
	100	36 ± 4.1	44 ± 3.6	39 ± 0.3	36 ± 2.1	39 ± 1.5	48 ± 6.4
	333	23 ± 0.6	46 ± 1.2	40 ± 2.2	34 ± 4.4	37 ± 5.5	45 ± 2.7[c]
	1,000	29 ± 3.0	39 ± 0.9		39 ± 5.4[c]	50 ± 5.0	47 ± 2.5[c]
	3,333	38 ± 5.9	43 ± 3.3	45 ± 3.6[c]	42 ± 4.1	55 ± 2.2[c]	45 ± 2.3[c]
	4,000	31 ± 4.6	39 ± 3.9		45 ± 1.5	61 ± 4.1[c]	46 ± 0.9[c]
Trial summary		Negative	Negative	Negative	Weakly positive	Weakly positive	Negative
Positive control		90 ± 4.4	78 ± 1.3	104 ± 3.6	92 ± 4.1	127 ± 11.5	158 ± 5.5

[a] The detailed protocol and these data are presented in Zeiger *et al.* (1988); 0 µg/plate dose is the solvent control.
[b] Revertants are presented as mean ± standard error from three plates.
[c] Precipitate on plate
[d] The positive controls in the absence of metabolic activation were sodium azide (TA100 and TA1535), 9-aminoacridine (TA97), and 4-nitro-*o*-phenylenediamine (TA98 and TA1538). The positive control for metabolic activation with all strains was 2-aminoanthracene.

TABLE C2
Induction of Sister Chromatid Exchanges in Chinese Hamster Ovary Cells by D&C Yellow No. 11 [a]

Compound	Dose (µg/mL)	Total Cells	No. of Chromo-somes	No. of SCEs	SCEs/ Chromo-some	SCEs/ Cell	Hrs in BrdU	Relative Change of SCEs/ Chromosome[b] (%)
−S9								
Trial 1								
Summary: Positive								
Dimethylsulfoxide		50	1,017	482	0.47	9.6	26.0	
Mitomycin-C	0.001	50	1,024	623	0.60	12.5	26.0	28.37
	0.010	5	102	216	2.11	43.2	26.0	346.82
D&C Yellow No. 11	0.27	50	1,029	579	0.56	11.6	26.0	18.72
	0.80	50	1,015	651	0.64	13.0	26.0	35.33*
	2.70	50	1,000	871	0.87	17.4	34.5[c]	83.78*
	8.00	0					26.0	
					P<0.001[d]			
Trial 2								
Summary: Positive								
Dimethylsulfoxide		50	1,030	507	0.49	10.1	26.0	
Mitomycin-C	0.001	50	1,014	629	0.62	12.6	26.0	26.02
	0.010	5	104	191	1.83	38.2	26.0	273.10
D&C Yellow No. 11	1.0	50	1,016	900	0.88	18.0	26.0	79.96*
	1.5	50	1,019	844	0.82	16.9	33.0[c]	68.27*
	2.7	25	504	522	1.03	20.9	33.0[c]	110.41*
	5.0	0					26.0	
					P<0.001			
+S9								
Summary: Positive								
Dimethylsulfoxide		50	1,022	457	0.44	9.1	26.0	
Cyclophosphamide	0.4	50	1,010	738	0.73	14.8	26.0	63.41
	2.0	5	103	245	2.37	49.0	26.0	431.94
D&C Yellow No. 11	2.7	50	1,012	532	0.52	10.6	26.0	17.56
	8.0	50	1,025	631	0.61	12.6	26.0	37.67*
	27.0[e]	50	1,028	1,055	1.02	21.1	34.5[c]	129.51*
	80.0[e]	0					26.0	
					P<0.001			

* Positive response (P<0.01)
[a] Study performed at Litton Bionetics, Inc. A detailed description of the protocol is presented in Galloway *et al*. (1987). SCE=sister chromatid exchange; BrdU=bromodeoxyuridine
[b] SCEs/chromosome in treated cells versus SCEs/chromosome in solvent control cells
[c] Because D&C Yellow No. 11 induced a delay in the cell division cycle, harvest time was extended to maximize the number of second- division metaphase cells available for analysis.
[d] Significance of relative SCEs/chromosome tested by the linear regression trend test versus log of the dose
[e] Color change (yellow); pH=7

TABLE C3
Induction of Chromosomal Aberrations in Chinese Hamster Ovary Cells by D&C Yellow No. 11 [a]

−S9					+S9				
Dose (µg/mL)	Total Cells	No. of Abs	Abs/ Cell	Cells with Abs (%)	Dose (µg/mL)	Total Cells	No. of Abs	Abs/ Cell	Cells with Abs (%)
Trial 1 - Harvest time: 10.5 hours Summary: Positive					Harvest time: 21.5 hours[b] Summary: Positive				
Dimethylsulfoxide					Dimethylsulfoxide				
	200	3	0.02	1.0		200	1	0.01	0.5
Mitomycin-C					Cyclophosphamide				
0.15	200	32	0.16	9.5	6.25	200	32	0.16	12.0
0.50	25	10	0.40	28.0	12.50	25	44	1.76	68.0
D&C Yellow No. 11					D&C Yellow No. 11				
5.0	200	37	0.19	16.0*	60.0	25	92	3.68	88.0*
7.5	50[c]	12	0.24	20.0*	69.7	25	109	4.36	80.0*
10.0	100[c]	18	0.18	13.0*	80.0	25	86	3.44	76.0*
15.0	0								
				P≤0.001[d]					P≤0.001
Trial 2 - Harvest time: 10.5 hours Summary: Positive									
Dimethylsulfoxide									
	200	4	0.02	1.5					
Mitomycin-C									
0.15	200	24	0.12	9.5					
0.50	25	9	0.36	24.0					
D&C Yellow No. 11									
10.0	25	16	0.64	40.0*					
12.5	25	18	0.72	40.0*					
15.0	50	28	0.56	40.0*					
				P≤0.001					

* Positive (P≤0.05)
[a] Study performed at Litton Bionetics, Inc. The detailed description of the protocol is presented in Galloway *et al*. (1987). Abs=aberrations
[b] Because of significant chemical-induced cell cycle delay, incubation time prior to addition of Colcemid was lengthened to provide sufficient first-division metaphase cells at harvest.
[c] Less than 200 cells scored due to lack of readable cells
[d] Significance of percent cells with aberrations tested by the linear regression trend test versus log of the dose

TABLE C4
Frequency of Micronuclei in Mouse Peripheral Blood Erythrocytes Following Treatment with D&C Yellow No. 11 in Feed for 13 Weeks [a]

Dose (ppm)	Micronucleated Normochromatic Erythrocytes/1,000 Cells[b]	Number of Mice per Dose Group
Male		
0	1.73 ± 0.12	10
5,000	1.97 ± 0.17	10
17,000	1.58 ± 0.13	10
50,000	1.71 ± 0.15	10
	P=0.504[c]	
Female		
0	1.23 ± 0.18	10
5,000	1.28 ± 0.11	9
17,000	0.94 ± 0.12	9
50,000	1.21 ± 0.12	10
	P=0.848	

[a] Study performed at USDA Western Regional Center, CA. Smears were prepared from peripheral blood samples obtained at the termination of a 13-week toxicity study on D&C Yellow No. 11 (NTP, 1991a).
[b] At least 10,000 normochromatic erythrocytes (NCEs) were scored per animal. Data are presented as mean ± standard error.
[c] Significance of micronucleated NCEs determined by analysis of variance

APPENDIX D
ORGAN WEIGHTS AND
ORGAN-WEIGHT-TO-BODY-WEIGHT RATIOS

TABLE D1 Organ Weights and Organ-Weight-to-Body-Weight Ratios for Rats at the 12-Month Interim Evaluation in the 2-Year Feed Study of D&C Yellow No. 11 154

TABLE D1
Organ Weights and Organ-Weight-to-Body-Weight Ratios for Rats at the 12-Month Interim Evaluation in the 2-Year Feed Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Male				
n	10	9	9	6
Necropsy body wt	471 ± 10	461 ± 10	446 ± 6	440 ± 13
R. Kidney				
Absolute	1.655 ± 0.042	1.680 ± 0.046	1.589 ± 0.045	1.620 ± 0.065
Relative	3.52 ± 0.05	3.65 ± 0.06	3.56 ± 0.08	3.68 ± 0.07
Liver				
Absolute	16.916 ± 0.599	19.417 ± 0.748**	19.599 ± 0.457**	20.480 ± 0.750**
Relative	35.89 ± 0.75	42.03 ± 0.94**	43.95 ± 1.12**	46.49 ± 0.75**
Female				
n	10	9	10	9
Necropsy body wt	267 ± 8	262 ± 5	257 ± 2	252 ± 6
R. Kidney				
Absolute	0.963 ± 0.021	0.912 ± 0.020	0.920 ± 0.027	0.930 ± 0.022
Relative	3.62 ± 0.08	3.48 ± 0.05	3.58 ± 0.09	3.69 ± 0.04
Liver				
Absolute	8.853 ± 0.204	9.656 ± 0.202*	10.566 ± 0.204**	10.780 ± 0.245**
Relative	33.21 ± 0.58	36.87 ± 0.44**	41.06 ± 0.56**	42.82 ± 0.50**

* Significantly different ($P \leq 0.05$) from the control group by Williams' or Dunnett's test
** $P \leq 0.01$
[a] Organ weights (absolute weights) and body weights are given in grams; organ-weight-to-body-weight ratios (relative weights) are given as mg organ weight/g body weight (mean ± standard error).

APPENDIX E
HEMATOLOGY RESULTS

TABLE E1 Hematology Data for Rats at the 12-Month Interim Evaluation in the 2-Year Feed Study of D&C Yellow No. 11 156

TABLE E1
Hematology Data for Rats at the 12-Month Interim Evaluation in the 2-Year Feed Study of D&C Yellow No. 11[a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Male				
n	10	9	9	6
Hematocrit (%)	45.4 ± 0.4	43.6 ± 0.6*	43.4 ± 0.5**	43.0 ± 0.6**
Hemoglobin (g/dL)	15.3 ± 0.2	14.7 ± 0.2*	14.3 ± 0.2**	14.5 ± 0.3**
Erythrocytes (10^6/µL)	8.76 ± 0.09	8.35 ± 0.09**	8.22 ± 0.09**	8.30 ± 0.16**
Reticulocytes (10^6/µL)	0.22 ± 0.02	0.24 ± 0.03	0.28 ± 0.02	0.23 ± 0.03
Nucleated erythrocytes (10^3/µL)	0.06 ± 0.04	0.08 ± 0.02	0.01 ± 0.01	0.05 ± 0.03
Mean cell volume (fL)	51.9 ± 0.3	52.2 ± 0.3	52.8 ± 0.2	51.9 ± 0.4
Mean cell hemoglobin (pg)	17.4 ± 0.1	17.6 ± 0.1	17.4 ± 0.1	17.4 ± 0.1
Mean cell hemoglobin concentration (g/dL)	33.6 ± 0.3	33.7 ± 0.2	33.0 ± 0.1	33.6 ± 0.4
Platelets (10^3/µL)	803.5 ± 20.7	889.6 ± 16.0*	897.9 ± 14.8**	885.7 ± 54.1*
Leukocytes (10^3/µL)	10.53 ± 1.09	8.63 ± 0.82	10.25 ± 0.55	8.89 ± 1.01
Segmented neutrophils (10^3/µL)	3.20 ± 0.61	2.66 ± 0.53	2.88 ± 0.22	2.55 ± 0.52
Lymphocytes (10^3/µL)	6.78 ± 0.52	5.66 ± 0.36	6.76 ± 0.53	6.01 ± 0.51
Monocytes (10^3/µL)	0.38 ± 0.08	0.19 ± 0.06	0.47 ± 0.08	0.24 ± 0.08
Eosinophils (10^3/µL)	0.16 ± 0.04	0.12 ± 0.04	0.13 ± 0.03	0.09 ± 0.03
Female				
n	10	9	10	9
Hematocrit (%)	45.5 ± 0.4	45.6 ± 0.5	44.9 ± 0.4	44.7 ± 0.5
Hemoglobin (g/dL)	15.0 ± 0.1	15.0 ± 0.1	14.8 ± 0.1	14.7 ± 0.2
Erythrocytes (10^6/µL)	7.97 ± 0.07	8.06 ± 0.06	7.90 ± 0.07	7.86 ± 0.15
Reticulocytes (10^6/µL)	0.19 ± 0.02	0.18 ± 0.02	0.19 ± 0.02	0.21 ± 0.03
Nucleated erythrocytes (10^3/µL)	0.10 ± 0.04	0.09 ± 0.04	0.08 ± 0.03[b]	0.05 ± 0.02
Mean cell volume (fL)	57.1 ± 0.4	56.5 ± 0.4	56.9 ± 0.3	57.0 ± 0.9
Mean cell hemoglobin (pg)	18.8 ± 0.1	18.7 ± 0.1	18.7 ± 0.1	18.7 ± 0.1
Mean cell hemoglobin concentration (g/dL)	32.9 ± 0.2	33.0 ± 0.2	32.8 ± 0.1	32.8 ± 0.3
Platelets (10^3/µL)	795.5 ± 67.3	767.7 ± 11.0	835.3 ± 65.1	794.7 ± 30.1
Leukocytes (10^3/µL)	6.41 ± 0.69	5.76 ± 0.60	4.79 ± 0.19[b]	5.41 ± 0.55
Segmented neutrophils (10^3/µL)	1.31 ± 0.24	1.13 ± 0.16	0.79 ± 0.08[b]	1.12 ± 0.20
Lymphocytes (10^3/µL)	4.79 ± 0.50	4.43 ± 0.49	3.81 ± 0.19[b]	4.04 ± 0.38
Monocytes (10^3/µL)	0.23 ± 0.07	0.16 ± 0.04	0.17 ± 0.04[b]	0.21 ± 0.07
Eosinophils (10^3/µL)	0.09 ± 0.02	0.04 ± 0.01	0.03 ± 0.02[b]	0.04 ± 0.02

* Significantly different ($P \leq 0.05$) from the control group by Dunn's or Shirley's test
** $P \leq 0.01$
[a] Mean ± standard error. Statistical tests were performed on unrounded data.
[b] n=9

APPENDIX F
REPRODUCTIVE TOXICITY STUDY RESULTS

TABLE F1 **Body Weight Gains in F_0 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11** **158**

TABLE F2 **Precohabitation Feed Consumption by F_0 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11** **159**

TABLE F3 **Maternal Toxicity in F_0 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11** **160**

TABLE F4 **Developmental Toxicity in F_1 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11** **161**

TABLE F1
Body Weight Gains in F_0 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
n	60	60	60	60
Male				
Days				
1 to 10	55.4 ± 0.5	52.7 ± 0.6**	50.6 ± 0.7**	48.8 ± 0.7**
10 to 16	23.6 ± 0.7	24.6 ± 0.7	20.1 ± 0.9**	21.2 ± 0.7**
16 to 22	31.1 ± 0.6	29.3 ± 0.5	29.2 ± 0.6	29.8 ± 0.5
22 to 29	21.5 ± 0.7	16.2 ± 2.3**	24.0 ± 0.5	21.7 ± 0.5
29 to 36	12.1 ± 0.5	16.7 ± 1.7**	13.9 ± 0.6	11.7 ± 0.5
36 to 43	19.3 ± 0.5	15.7 ± 0.6**	13.7 ± 0.6**	15.2 ± 0.4**
43 to 50	18.9 ± 0.4	18.2 ± 0.4	17.9 ± 0.4	15.9 ± 0.4**
50 to 57	9.7 ± 0.6	12.6 ± 0.7**	12.0 ± 0.4*	10.6 ± 0.4
57 to 64	8.0 ± 0.6	6.3 ± 0.7	6.8 ± 0.5	3.4 ± 1.7**
64 to 71	9.7 ± 0.4	6.7 ± 0.5	7.2 ± 0.4	12.3 ± 2.4
78 to 85	7.2 ± 0.6	10.7 ± 2.3	6.4 ± 0.5	6.7 ± 1.2
1 to 71	209.3 ± 1.7	199.0 ± 1.6**	195.3 ± 1.6**	190.5 ± 2.0**
1 to 85	228.6 ± 2.0	218.1 ± 2.0**	213.0 ± 1.8**	206.0 ± 1.7**
Female				
Days				
1 to 3	10.1 ± 0.3[b]	9.7 ± 0.5	8.8 ± 0.2**	8.1 ± 0.2**
3 to 11	20.4 ± 0.5	20.1 ± 0.5	20.3 ± 0.4	21.0 ± 0.4
11 to 17	10.8 ± 0.4	10.6 ± 0.4	10.8 ± 0.4	9.6 ± 0.4
17 to 24	7.9 ± 0.3	7.1 ± 0.4	7.5 ± 0.3	7.2 ± 0.3
24 to 31	9.9 ± 0.4	9.7 ± 0.3	9.1 ± 0.3	10.1 ± 0.4
31 to 38	7.7 ± 0.4	7.5 ± 0.3	7.5 ± 0.4	6.2 ± 0.3*
38 to 45	5.1 ± 0.4	4.6 ± 0.3	4.7 ± 0.4	4.9 ± 0.5
45 to 52	6.5 ± 0.4	6.3 ± 0.3	5.5 ± 0.3*	5.2 ± 0.4**
52 to 59	2.0 ± 0.3	3.0 ± 0.4	3.8 ± 0.4**	3.1 ± 0.3**
59 to 66	5.2 ± 0.4	5.9 ± 0.4	5.6 ± 0.4	5.3 ± 0.3
1 to 66	85.6 ± 1.1[b]	84.3 ± 0.9	83.4 ± 1.1	80.5 ± 1.0**

* Significantly different ($P \leq 0.05$) from the control group by Williams' or Dunnett's test
** $P \leq 0.01$
[a] Body weight gains are given in grams (mean ± standard error).
[b] n=59

TABLE F2
Precohabitation Feed Consumption by F_0 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Number of cages[b]	12	12	12	12
Male				
Days				
2 to 9	17.0 ± 0.2	16.3 ± 0.5	15.8 ± 0.3**	16.2 ± 0.4
16 to 23	17.0 ± 0.1	17.0 ± 0.1	16.4 ± 0.1**	16.8 ± 0.1
23 to 30	18.0 ± 0.2	17.0 ± 0.6	17.2 ± 0.7*	17.3 ± 0.4
30 to 37	18.4 ± 0.5	18.0 ± 0.3	17.3 ± 0.9	16.8 ± 0.4*
37 to 44	18.7 ± 0.8	18.4 ± 0.2	18.3 ± 0.1	17.9 ± 0.1
44 to 51	18.8 ± 0.8	23.8 ± 0.6**	23.4 ± 0.5*	21.6 ± 1.1
51 to 58	15.8 ± 1.6	10.4 ± 1.9	11.2 ± 2.0	11.8 ± 2.0[c]
58 to 65	16.1 ± 0.3	16.4 ± 0.5	16.8 ± 0.5	17.0 ± 0.6
Female				
Days				
2 to 9	12.6 ± 0.1	12.1 ± 0.1*	12.1 ± 0.2*	12.3 ± 0.1
9 to 16	11.4 ± 0.1	11.1 ± 0.1	11.1 ± 0.1	11.2 ± 0.1
16 to 23	11.1 ± 0.1	11.1 ± 0.1	10.9 ± 0.1	11.0 ± 0.2
23 to 30	11.4 ± 0.7	12.2 ± 0.9	10.7 ± 0.3*	11.3 ± 0.1
30 to 37	11.6 ± 0.2	11.2 ± 0.1	11.2 ± 0.1	11.4 ± 0.2
37 to 44	11.7 ± 1.0	11.9 ± 0.2	12.0 ± 0.1	11.7 ± 0.1
44 to 51	10.7 ± 1.0	10.3 ± 0.4	10.3 ± 0.5	10.9 ± 0.1
51 to 58	12.0 ± 0.6	11.8 ± 0.8	11.4 ± 0.2	11.4 ± 0.3
58 to 65	10.5 ± 0.3	10.0 ± 0.4	10.4 ± 0.1	10.8 ± 0.3

* Significantly different ($P \leq 0.05$) from the control group by Dunn's or Shirley's test
** $P \leq 0.01$
[a] Feed consumption data are given in grams per animal per day (mean ± standard error).
[b] Five rats per cage
[c] Feed consumption was not measured for one cage.

TABLE F3
Maternal Toxicity in F_0 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Number examined	60	60	60	60
Number pregnant	43	39	49	46
Maternal body weight gains during gestation (g)				
Days				
0 to 6	9.1 ± 2.7	10.9 ± 4.0	10.9 ± 3.0	10.7 ± 3.7
6 to 15	32.9 ± 4.1	31.2 ± 5.1	28.8 ± 4.9**	30.4 ± 4.5*
15 to 21	47.0 ± 9.3	50.0 ± 7.5	47.6 ± 13.0	49.1 ± 7.3[b]
0 to 21	89.0 ± 10.8	92.2 ± 12.2	87.4 ± 17.0	90.4 ± 10.9[b]
Maternal body weight gains during lactation (g)				
Days				
1 to 4	–3.5 ± 6.8	–5.8 ± 8.0[c]	–7.7 ± 11.8[d]	–8.0 ± 9.4[e]
4 to 14	18.6 ± 12.9	13.9 ± 14.0[c]	18.1 ± 11.1[d]	15.5 ± 9.4[e]
14 to 21	–0.2 ± 16.2	1.9 ± 18.9	–1.6 ± 16.0[f]	2.4 ± 16.8[e]
1 to 21	14.9 ± 9.4	10.3 ± 13.6	8.9 ± 15.6[g]	9.9 ± 15.0[e]
Duration of gestation (days)[h]	23.0 ± 0.0	23.0 ± 0.2	23.1 ± 0.3	23.0 ± 0.0

* Significantly different ($P \leq 0.05$) from the control group by Dunnett's test
** $P \leq 0.01$
[a] Mean ± standard deviation
[b] n=44
[c] n=38
[d] n=46
[e] n=45
[f] n=48
[g] n=47
[h] Data for rats with confirmed mating dates

TABLE F4
Developmental Toxicity in F_1 Rats in the Reproductive Toxicity Study of D&C Yellow No. 11 [a]

	0 ppm	500 ppm	1,700 ppm	5,000 ppm
Number of litters examined	43	39	49	46
Pups delivered (total)	418	396	471	459
Pups delivered per litter	9.7 ± 2.5	10.2 ± 2.3	9.6 ± 3.3	10.0 ± 2.0
Percent male pups	50.0 ± 16.9	51.8 ± 18.5	48.2 ± 18.1	48.7 ± 20.2
Pups surviving 4 days (precull) per number of pups delivered	410/416 (99%)	388/396 (98%)	463/470 (99%)	447/458 (98%)
Pups surviving 21 days per number of pups selected on day 4 (postcull)	324/327 (99%)	297/299 (99%)	349/349 (100%)	347/350 (99%)
Pup weight per litter (g)				
Day				
1	5.23 ± 0.04	5.33 ± 0.05	5.37 ± 0.04*[b]	5.34 ± 0.03
4 (precull)	7.26 ± 0.08[c]	7.22 ± 0.12[d]	7.13 ± 0.13[e]	6.96 ± 0.07[f]
4 (postcull)	7.29 ± 0.08[c]	7.28 ± 0.12[d]	7.18 ± 0.12[e]	7.01 ± 0.06[f]
14	20.7 ± 0.2	19.6 ± 0.2**	19.5 ± 0.2**[b]	19.1 ± 0.2**[g]
21	30.8 ± 0.3	28.8 ± 0.3**	28.1 ± 0.2**[b]	27.1 ± 0.2**[g]

* Significantly different ($P \leq 0.05$) from the control group by Williams' or Dunnett's test
** $P \leq 0.01$
[a] Data are presented as mean ± standard deviation (pups delivered/litter) or mean ± standard error (pup weights/litter).
[b] n=48
[c] n=21
[d] n=23
[e] n=25
[f] n=28
[g] n=45

APPENDIX G
CHEMICAL CHARACTERIZATION AND DOSE FORMULATION STUDIES

PROCUREMENT AND CHARACTERIZATION OF D&C YELLOW NO. 11 164
PREPARATION AND ANALYSIS OF DOSE FORMULATIONS 165
FIGURE G1 Infrared Absorption Spectrum of D&C Yellow No. 11 166
FIGURE G2 Nuclear Magnetic Resonance Spectrum of D&C Yellow No. 11 167
TABLE G1 Preparation and Storage of Dose Formulations in the Feed Studies of D&C Yellow No. 11 168
TABLE G2 Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 169
TABLE G3 Results of Referee Analysis of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 174

CHEMICAL CHARACTERIZATION AND DOSE FORMULATION STUDIES

PROCUREMENT AND CHARACTERIZATION OF D&C YELLOW NO. 11

D&C Yellow No. 11 was obtained from H. Kohnstamm and Company, Inc. (New York), in one lot (ZB2016) and certified by the Food and Drug Administration, Division of Color Technology. Lot ZB2016 was used during the reproductive toxicity and 2-year studies. Identity, purity, and stability analyses were conducted by the analytical chemistry laboratory, Midwest Research Institute (Kansas City, MO). Reports on analyses performed in support of the D&C Yellow No. 11 studies are on file at the National Institute of Environmental Health Sciences.

Lot ZB2016, a yellow powder, was identified as D&C Yellow No. 11 by infrared, ultraviolet/visible, and nuclear magnetic resonance spectrometry. All spectra were consistent with those expected for the structure of D&C Yellow No. 11. However, the nuclear magnetic resonance spectrum indicated impurities. Direct inlet mass spectrometry confirmed the identity of the compound as D&C Yellow No. 11 and indicated the presence of a monochlorinated isomer. The infrared and nuclear magnetic resonance spectra are presented in Figures G1 and G2. The observed melting point range, 240.9 ° to 242.1 ° C, was consistent with the melting point range, 235 ° to 240 ° C, specified by the manufacturer for purified D&C Yellow No. 11.

The purity was determined by elemental analyses, Karl Fischer water analysis, thin-layer chromatography (TLC), and high-performance liquid chromatography (HPLC). TLC was performed on Silica Gel 60 F-254 plates with two solvent systems: 1) methylene chloride:acetone:glacial acetic acid (70:26:4) and 2) toluene:methanol (95:5). Quinoline was used as a reference standard. Plates were examined under visible and ultraviolet light (254 and 366 nm) and with iodine vapors. HPLC was performed with a Waters µBondapak C_{18} column using ultraviolet (280 nm) and visible (436 nm) detection and a solvent system of water:methanol (37:63) at a flow rate of 1.0 mL/min.

Elemental analyses for carbon, hydrogen, and nitrogen were in agreement with the theoretical values for D&C Yellow No. 11. Karl Fischer water analysis indicated less than 0.02% water. TLC indicated one major spot by system 1 and one major spot and one trace impurity by system 2. HPLC indicated a major peak and two impurities with areas greater than 0.1% relative to the major peak at both 280 and 436 nm. A high-speed scanning detector (Hewlett-Packard 1040A) was used in conjunction with HPLC with a solvent system of water:methanol (32:68) to obtain an ultraviolet/visible absorption spectrum for the largest of the two impurity peaks. The results indicated that this impurity was similar in structure to that of the major peak. The overall purity was determined to be approximately 99%.

Stability studies of the bulk chemical were performed using the HPLC system described for the purity analysis except with a solvent system ratio of 32:68, ultraviolet detection at 254 nm, and valerophenone as an internal standard. These studies indicated that D&C Yellow No. 11 was stable as a bulk chemical for 2 weeks when stored protected from light at temperatures up to 60 ° C. To ensure stability, the bulk chemical was stored in its original packaging (metal cans or cardboard drums) at room temperature protected from light. The stability of the bulk chemical was monitored by the study laboratory approximately every 4 months during the studies and within 30 days of the end of the 2-year study by HPLC. No degradation of the bulk chemical was observed.

PREPARATION AND ANALYSIS OF DOSE FORMULATIONS

The dose formulations were prepared every 2 weeks by mixing D&C Yellow No. 11 with feed (Table G1). A D&C Yellow No. 11/feed premix was made by hand and then blended with feed in a Patterson-Kelly twin-shell

blender for 15 minutes with the intensifier bar on for the first 5 minutes. During the studies, dose formulations were stored in double-thickness plastic bags in rigid plastic containers at room temperature protected from light for up to 3 weeks.

Homogeneity and stability studies of the 500 ppm dose formulation were performed by the analytical chemistry laboratory. Extracts were prepared by shaking 10 g samples with 100 mL of acetone in a wrist-action shaker for 15 minutes. After centrifugation, 5 mL aliquots of the extracts were diluted to 50 mL with a water:methanol solution (20:80) and filtered. HPLC was performed with a Brownlee RP-18 column using visible light detection and a mobile phase of water:methanol (20:80) at a flow rate of 1.0 mL/minute. Homogeneity was confirmed, and the dose formulations were determined to be stable for up to 3 weeks when stored protected from light at room temperature and for 7 days when stored open to air and light.

Periodic analyses of the dose formulations of D&C Yellow No. 11 were conducted at the study laboratory using visible spectrometry. Dose formulations were analyzed approximately every 8 to 10 weeks (Table G2). All dose formulations used in the studies were within 10% of the target concentrations. One formulation was remixed due to an unacceptable ratio of duplicate analyses. The remix was within acceptable limits and the original mix was not used for dosing. Results of a referee analysis performed by the analytical chemistry laboratory indicated good agreement with the results obtained by the study laboratory (Table G3).

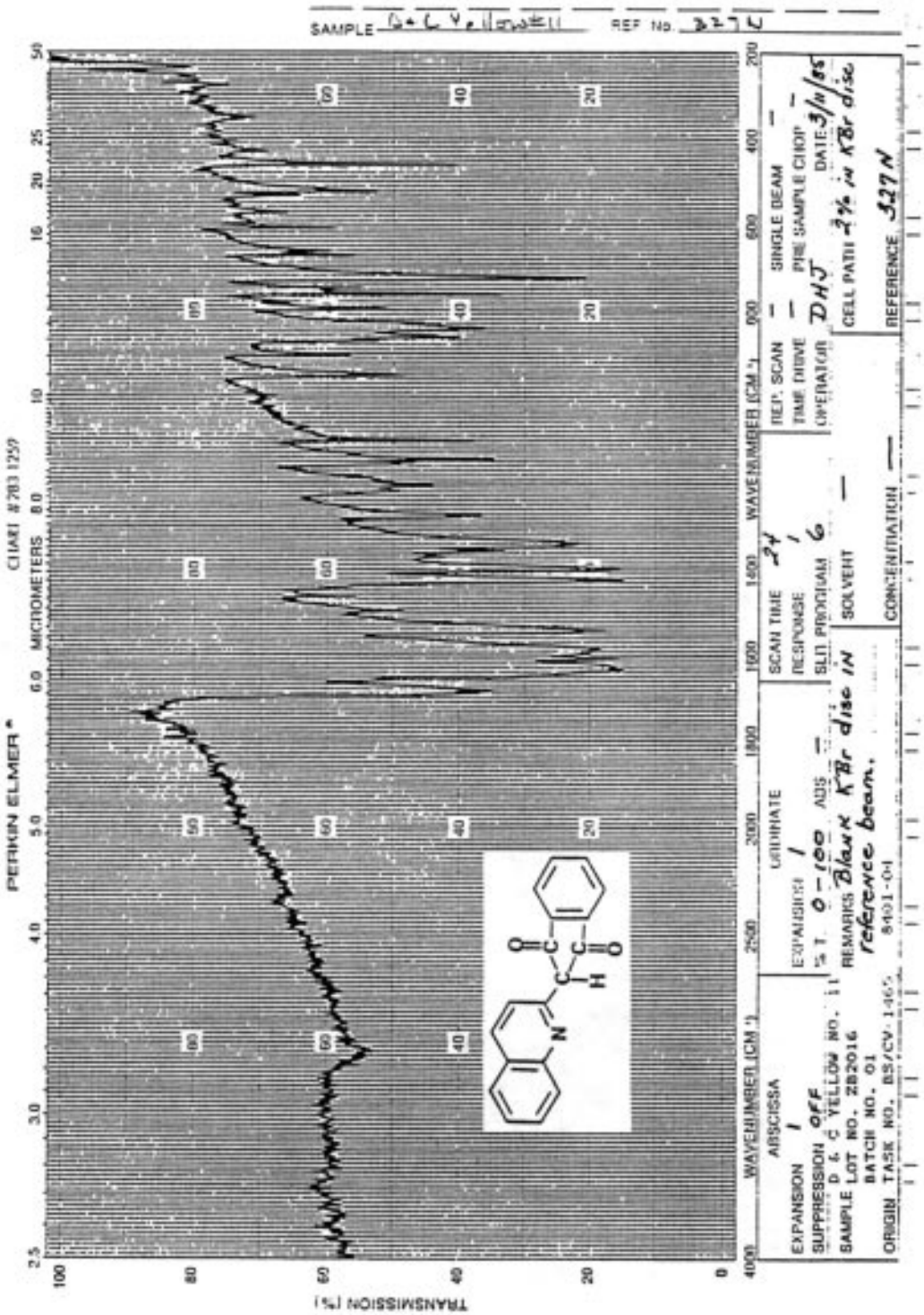


FIGURE G1
Infrared Absorption Spectrum of D&C Yellow No. 11

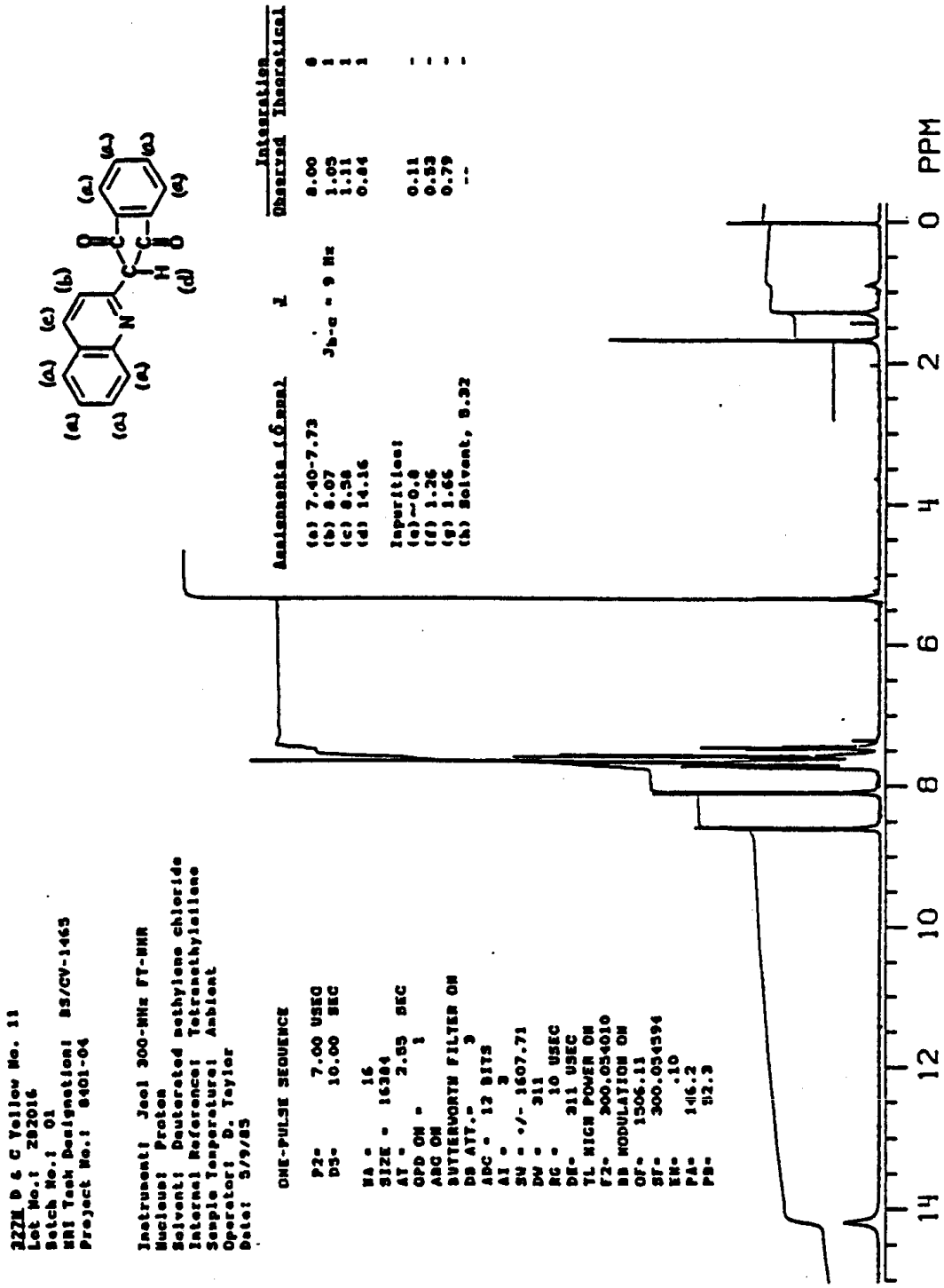


FIGURE G2
Nuclear Magnetic Resonance Spectrum of D&C Yellow No. 11

TABLE G1
Preparation and Storage of Dose Formulations in the Feed Studies of D&C Yellow No. 11

Reproductive Toxicity Study	2-Year Study
Preparation A premix of feed and D&C Yellow No. 11 was prepared, then layered into the remaining feed and blended in a Patterson-Kelly twin-shell blender with the intensifier bar on for 5 minutes and off for 10 minutes. Doses were prepared every 2 weeks.	Same as reproductive toxicity study
Chemical Lot Number ZB2016	ZB2016
Maximum Storage Time 3 weeks	3 weeks
Storage Conditions Stored in double-thickness plastic bags in rigid plastic containers at room temperature in the dark.	Same as reproductive toxicity study
Study Laboratory Southern Research Institute (Birmingham, AL)	Southern Research Institute (Birmingham, AL)
Referee Laboratory Midwest Research Institute (Kansas City, MO)	Midwest Research Institute (Kansas City, MO)

TABLE G2
Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11

Date Prepared	Date Analyzed	Target Concentration (ppm)	Determined Concentration[a] (ppm)	Difference from Target (%)
27 November 1989[b]	27–28 November 1989	500	483[c]	–3
		500	486[d]	–3
		500	483[e]	–3
		5,000	5,050[c]	+1
		5,000	4,910[d]	–2
		5,000	4,980[e]	0
12 December 1989	13 December 1989	500	480	–4
		500	489	–2
		500	486	–3
		1,700	1,700	0
		1,700	1,680	–1
		1,700	1,700	0
		5,000	5,040	+1
		5,000	4,980	0
		5,000	5,030	+1
20 February 1990	20–21 February 1990	500	475	–5
		500	472	–6
		500	484	–3
		500	481	–4
		1,700	1,680	–1
		1,700	1,680	–1
		1,700	1,710	+1
		1,700	1,680	–1
		5,000	4,990	0
		5,000	5,100	+2
		5,000	5,070	+1
		5,000	4,980	0
17 April 1990	18 April 1990	500	473	–5
		500	454	–9
		500	470	–6
		500	478	–4
		1,700	1,630	–4
		1,700	1,600	–6
		1,700	1,580	–7
		1,700	1,640	–4
		5,000	4,780	–4
		5,000	4,880	–2
		5,000	4,780	–4
		5,000	4,880	–2

TABLE G2
Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 (continued)

Date Prepared	Date Analyzed	Target Concentration (ppm)	Determined Concentration (ppm)	Difference from Target (%)
12 June 1990	13–14 June 1990	500	471	–6
		500	468	–6
		500	487	–3
		500	461	–8
		1,700	1,650	–3
		1,700	1,630	–4
		1,700	1,540	–9
		1,700	1,550	–9
		5,000	4,750	–5
		5,000	4,520	–10
		5,000	4,610	–8
		5,000	4,970	–1
	6 July 1990[f]	500	431	–14
		1,700	1,620	–5
		5,000	4,760	–5
21 August 1990	21 August 1990	500	503	+1
		500	494	–1
		500	519	+4
		500	500	0
		1,700	1,680	–1
		1,700	1,700	0
		1,700	1,680	–1
		1,700	1,670	–2
		5,000	4,910	–2
		5,000	4,930	–1
		5,000	4,970	–1
		5,000	5,060	+1
16 October 1990	16–17 October 1990	500	501	0
		500	495	–1
		500	488	–2
		500	488	–2
		1,700	1,670	–2
		1,700	1,670	–2
		1,700	1,660	–2
		1,700	1,710	+1
		5,000	4,970	–1
		5,000	4,890	–2
		5,000	4,960	–1
		5,000	4,950	–1

TABLE G2
Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 (continued)

Date Prepared	Date Analyzed	Target Concentration (ppm)	Determined Concentration (ppm)	Difference from Target (%)
11 December 1990	12 December 1990	500	491	–2
		500	494	–1
		500	494	–1
		500	500	0
		1,700	1,720	+1
		1,700	1,690	–1
		1,700	1,700	0
		1,700	1,690	–1
		5,000	5,000	0
		5,000	5,060	+1
		5,000	5,020	0
		5,000	5,010	0
	7 January 1991[f]	500	477	–5
		1,700	1,650	–3
		5,000	4,910	–2
5 February 1991	5–7 February 1991	500	495	–1
		500	501	0
		500	495	–1
		500	526	+5
		1,700	1,670	–2
		1,700	1,810	+6
		1,700	1,680	–1
		1,700	1,670	–2
		5,000	4,940	–1
		5,000	4,840	–3
		5,000	4,900	–2
		5,000	4,840	–3
2 April 1991	3–4 April 1991	500	469	–6
		500	466	–7
		500	473	–5
		500	469	–6
		1,700	1,700	0
		1,700	1,700	0
		1,700	1,720	+1
		1,700	1,710	+1
		5,000	5,130	+3
		5,000	5,310	+6
		5,000	5,100	+2
		5,000	5,110	+2

TABLE G2
Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 (continued)

Date Prepared	Date Analyzed	Target Concentration (ppm)	Determined Concentration (ppm)	Difference from Target (%)
11 June 1991	11–12 June 1991	500	484	–3
		500	496	–1
		500	493	–1
		500	487	–3
		1,700	1,680	–1
		1,700	1,680	–1
		1,700	1,670	–2
		1,700	1,680	–1
		5,000	4,940	–1
		5,000	4,960	–1
		5,000	4,970	–1
		5,000	4,970	–1
	25 and 27 June 1991[f]	500	461	–8
		500	480	–4
		500	471	–6
		500	464	–7
		1,700	1,640	–4
		1,700	1,640	–4
		1,700	1,630	–4
		1,700	1,660	–2
		5,000	4,860	–3
		5,000	4,860	–3
		5,000	4,940	–1
		5,000	4,930	–1
20 August 1991	20–21 August 1991	500	505	+1
		500	512	+2
		500	509	+2
		500	509	+2
		1,700	1,710	+1
		1,700	1,700	0
		1,700	1,700	0
		1,700	1,700	0
		5,000	4,940	–1
		5,000	4,960	–1
		5,000	5,080	+2
		5,000	4,970	–1
15 October 1991	16–17 October 1991	500	479	–4
		500	482	–4
		500	488	–2
		500	498	0
		1,700	1,680	–1
		1,700	1,680	–1
		1,700	1,650	–3
		1,700	1,680	–1
		5,000	4,860	–3
		5,000	4,850	–3
		5,000	4,940	–1
		5,000	4,910	–2

TABLE G2
Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 (continued)

Date Prepared	Date Analyzed	Target Concentration (ppm)	Determined Concentration (ppm)	Difference from Target (%)
10 December 1991	10–11 December 1991	500	493	–1
		500	502	0
		500	505	+1
		500	489	–2
		1,700	1,670	–2
		1,700	1,650	–3
		1,700	1,670	–2
		1,700	1,680	–1
		5,000	4,920	–2
		5,000	4,870	–3
		5,000	5,080	+2
		5,000	4,930	–1
	31 December 1991 – 2–3 January 1992[f]	500	480	–4
		500	480	–4
		500	470	–6
		1,700	1,650	–3
		1,700	1,680	–1
		1,700	1,650	–3
		5,000	4,890	–2
		5,000	4,840	–3
		5,000	4,940	–1
4 February 1992	4–5 February 1992	500	501	0
		500	517	+3
		500	501	0
		500	494	–1
		1,700	1,700	0
		1,700	1,780[g]	+5
		1,700	1,690	–1
		1,700	1,690	–1
		5,000	4,940	–1
		5,000	4,930	–1
		5,000	5,020	0
		5,000	4,930	–1
6 February 1992	7 February 1992	1,700	1,690[h]	–1

TABLE G2
Results of Analyses of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11 (continued)

Date Prepared	Date Analyzed	Target Concentration (ppm)	Determined Concentration (ppm)	Difference from Target (%)
31 March 1992	31 March 1992 – 1 April 1992	500	493	–1
		500	499	0
		500	502	0
		1,700	1,690	–1
		1,700	1,670	–2
		1,700	1,680	–1
		5,000	4,920	–2
		5,000	5,050	+1
		5,000	4,870	–3

[a] Results of duplicate analyses
[b] Homogeneity analyses, formulations not used for dosing
[c] Sample from top right of twin-shell blender
[d] Sample from top left of twin-shell blender
[e] Sample from bottom of twin-shell blender
[f] Animal room samples
[g] Not used for dosing due to unacceptable ratio of duplicate analyses (0.89)
[h] Result of remix

TABLE G3
Results of Referee Analysis of Dose Formulations Administered to Rats in the Reproductive Toxicity and 2-Year Feed Studies of D&C Yellow No. 11

Date Prepared (ppm)	Determined Concentration (ppm) Target Concentration Laboratory[a]	Study Laboratory[b]	Referee
12 December 1989	500	480	500 ± 2

[a] Results of duplicate analyses
[b] Results of triplicate analyses (mean ± standard error)

APPENDIX H
FEED AND COMPOUND CONSUMPTION IN THE 2-YEAR FEED STUDY OF D&C YELLOW NO. 11

TABLE H1 Feed and Compound Consumption by Male Rats in the 2-Year Feed Study of D&C Yellow No. 11 176
TABLE H2 Feed and Compound Consumption by Female Rats in the 2-Year Feed Study of D&C Yellow No. 11 177

TABLE H1
Feed and Compound Consumption by Male Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm		500 ppm			1,700 ppm			5,000 ppm		
Week	Feed (g/day)[a]	Body Weight (g)	Feed (g/day)	Body Weight (g)	Dose/ Day[b] (mg/kg)	Feed (g/day)	Body Weight (g)	Dose/ Day (mg/kg)	Feed (g/day)	Body Weight (g)	Dose/ Day (mg/kg)
2	15.1	159	14.3	145	50	13.9	142	167	13.5	133	506
6	18.1	277	18.6	260	36	18.5	253	125	17.5	240	363
10	18.1	327	17.9	315	28	18.0	310	99	18.2	297	308
13	16.7	353	17.2	343	25	16.8	337	85	17.1	323	265
17	17.2	383	16.7	371	23	17.2	365	80	16.9	352	240
21	17.0	401	16.3	390	21	17.1	385	76	16.7	370	226
25	16.1	416	16.5	405	20	15.7	400	67	16.7	384	217
29	16.7	431	17.1	421	20	17.1	413	70	17.6	400	220
33	14.9	425	15.0	427	18	15.9	423	64	16.7	404	206
37	16.5	449	17.8	436	20	17.5	432	69	16.8	416	202
41	16.9	455	17.0	443	19	17.2	439	66	17.4	421	207
45	17.5	456	16.8	449	19	17.2	443	66	16.7	423	197
49	16.4	464	16.9	454	19	17.0	450	64	16.9	430	197
53	16.5	472	16.6	460	18	15.9	457	59	17.3	438	198
57	17.1	472	16.6	461	18	16.8	460	62	17.3	438	197
61	16.7	475	16.1	462	17	16.3	461	60	16.5	441	187
65	15.9	477	15.7	463	17	15.9	460	59	16.3	439	185
69	15.7	475	15.8	463	17	16.3	462	60	16.6	438	189
73	16.1	472	15.2	460	17	16.4	456	61	16.5	436	189
77	15.7	473	15.4	451	17	16.7	451	63	17.1	433	197
81	16.1	470	15.4	446	17	15.8	442	61	16.1	425	190
85	14.2	463	14.8	445	17	14.9	435	58	15.3	413	185
89	14.3	452	14.0	439	16	15.6	433	61	15.2	404	188
93	14.3	451	14.1	425	17	15.8	422	64	15.1	401	188
97	12.7	446	14.9	425	17	13.3	420	54	13.6	387	175
101	13.6	435	14.2	414	17	16.5	403	70	15.3	368	208
Means for weeks											
1-13	17.0	279	17.0	265	35	16.8	260	119	16.6	248	360
14-52	16.6	431	16.7	422	20	16.9	417	69	16.9	400	212
53-101	15.3	464	15.3	447	17	15.9	443	61	16.0	420	191

[a] Grams of feed consumed per animal per day
[b] Milligrams of D&C Yellow No. 11 consumed per kilogram body weight per day

TABLE H2
Feed and Compound Consumption by Female Rats in the 2-Year Feed Study of D&C Yellow No. 11

	0 ppm		500 ppm			1,700 ppm			5,000 ppm		
Week	Feed (g/day)[a]	Body Weight (g)	Feed (g/day)	Body Weight (g)	Dose/ Day[b] (mg/kg)	Feed (g/day)	Body Weight (g)	Dose/ Day (mg/kg)	Feed (g/day)	Body Weight (g)	Dose/ Day (mg/kg)
2	12.1	126	11.0	117	47	10.9	115	161	10.9	113	484
6	12.1	168	12.1	161	38	11.3	156	122	11.6	155	376
10	11.5	187	11.4	179	32	11.0	173	108	11.0	174	315
16	10.7	206	10.7	200	27	10.2	196	88	10.5	194	272
21	10.4	212	10.7	205	26	10.2	201	86	10.8	197	274
24	10.4	217	10.7	212	25	10.2	207	84	10.8	205	263
28	11.0	227	11.0	218	25	10.3	215	81	10.7	211	253
32	10.2	233	10.9	227	24	10.5	221	80	11.0	219	253
36	9.9	238	10.4	231	23	10.0	222	77	10.1	223	226
40	11.2	246	11.4	240	24	11.7	231	86	12.3	233	264
44	11.6	255	11.1	248	22	11.0	238	79	11.4	240	238
48	12.3	265	12.2	258	24	11.9	251	80	12.1	250	241
52	11.9	277	12.3	276	22	11.7	259	76	11.7	263	223
56	12.5	290	12.2	285	21	12.1	273	75	12.2	275	221
60	12.1	296	12.9	293	22	12.1	280	73	12.5	282	221
64	12.8	307	12.1	303	20	12.1	289	71	11.7	291	201
68	12.6	314	12.2	312	20	12.4	300	70	12.3	299	205
72	12.4	320	12.7	317	20	12.6	304	70	13.4	307	219
76	12.2	323	12.6	320	20	12.2	305	68	12.8	308	208
80	12.9	331	12.6	323	20	12.6	310	69	12.6	313	201
84	12.8	339	12.8	330	19	12.2	315	66	11.6	317	183
88	12.0	342	12.1	330	18	12.5	320	66	12.5	319	196
92	12.4	348	11.5	333	17	12.6	327	65	12.6	320	197
96	13.2	357	13.0	342	19	12.6	331	65	12.2	332	184
100	12.2	355	12.7	344	18	12.6	336	64	12.5	334	188
104	11.6	354	12.4	349	18	12.2	337	61	12.2	333	183
Means for weeks											
1-13	11.9	160	11.5	152	39	11.0	148	131	11.2	147	392
14-52	11.0	238	11.1	231	24	10.8	224	82	11.1	224	251
53-104	12.4	329	12.5	322	19	12.4	310	68	12.4	310	201

[a] Grams of feed consumed per animal per day
[b] Milligrams of D&C Yellow No. 11 consumed per kilogram body weight per day

APPENDIX I
INGREDIENTS, NUTRIENT COMPOSITION, AND CONTAMINANT LEVELS IN NIH-07 RAT AND MOUSE RATION

TABLE I1	Ingredients of NIH-07 Rat and Mouse Ration	180
TABLE I2	Vitamins and Minerals in NIH-07 Rat and Mouse Ration	180
TABLE I3	Nutrient Composition of NIH-07 Rat and Mouse Ration	181
TABLE I4	Contaminant Levels in NIH-07 Rat and Mouse Ration	182

TABLE I1
Ingredients of NIH-07 Rat and Mouse Ration[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
Dried brewer's 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] NCI, 1976; NIH, 1978
[b] Ingredients were ground to pass through a U.S. Standard Screen No. 16 before being mixed.

TABLE I2
Vitamins and Minerals in NIH-07 Rat and Mouse Ration[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
K_3	2.8 g	Menadione
d-α-Tocopheryl acetate	20,000 IU	
Choline	560.0 g	Choline chloride
Folic acid	2.2 g	
Niacin	30.0 g	
d-Pantothenic acid	18.0 g	*d*-Calcium pantothenate
Riboflavin	3.4 g	
Thiamine	10.0 g	Thiamine mononitrate
B_{12}	4,000 µg	
Pyridoxine	1.7 g	Pyridoxine hydrochloride
Biotin	140.0 mg	*d*-Biotin
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 I3
Nutrient Composition of NIH-07 Rat and Mouse Ration

Nutrient	Mean ± Standard Deviation	Range	Number of Samples
Protein (% by weight)	23.68 ± 0.54	22.5 – 25.2	27
Crude fat (% by weight)	5.25 ± 0.23	4.80 – 5.80	27
Crude fiber (% by weight)	3.56 ± 0.43	2.60 – 4.30	27
Ash (% by weight)	6.48 ± 0.19	6.12 – 6.97	27
Amino Acids (% of total diet)			
Arginine	1.280 ± 0.083	1.110 – 1.390	11
Cystine	0.308 ± 0.071	0.181 – 0.400	11
Glycine	1.158 ± 0.048	1.060 – 1.220	11
Histidine	0.584 ± 0.027	0.531 – 0.630	11
Isoleucine	0.917 ± 0.033	0.867 – 0.965	11
Leucine	1.975 ± 0.051	1.850 – 2.040	11
Lysine	1.274 ± 0.049	1.200 – 1.370	11
Methionine	0.437 ± 0.109	0.306 – 0.699	11
Phenylalanine	0.999 ± 0.120	0.665 – 1.110	11
Threonine	0.904 ± 0.058	0.824 – 0.985	11
Tryptophan	0.218 ± 0.153	0.107 – 0.671	11
Tyrosine	0.685 ± 0.094	0.564 – 0.794	11
Valine	1.086 ± 0.055	0.962 – 1.170	11
Essential Fatty Acids (% of total diet)			
Linoleic	2.407 ± 0.227	1.830 – 2.570	10
Linolenic	0.259 ± 0.065	0.100 – 0.320	10
Vitamins			
Vitamin A (IU/kg)	6,821 ± 1,531	4,290 – 12,540	27
Vitamin D (IU/kg)	4,450 ± 1,382	3,000 – 6,300	4
α-Tocopherol (ppm)	36.12 ± 9.15	22.50 – 48.9	10
Thiamine (ppm)	18.81 ± 2.11	15.0 – 25.0	27
Riboflavin (ppm)	7.83 ± 0.923	6.10 – 9.00	11
Niacin (ppm)	98.64 ± 25.5	65.0 – 150.0	10
Pantothenic acid (ppm)	30.55 ± 3.52	23.0 – 34.6	11
Pyridoxine (ppm)	9.11 ± 2.53	5.60 – 14.0	11
Folic acid (ppm)	2.46 ± 0.63	1.80 – 3.70	11
Biotin (ppm)	0.268 ± 0.047	0.190 – 0.354	11
Vitamin B_{12} (ppb)	40.5 ± 19.1	10.6 – 65.0	11
Choline (ppm)	2,991 ± 382	2,300 – 3,430	10
Minerals			
Calcium (%)	1.18 ± 0.09	1.02 – 1.37	27
Phosphorus (%)	0.94 ± 0.046	0.800 – 1.03	27
Potassium (%)	0.886 ± 0.063	0.772 – 0.971	9
Chloride (%)	0.529 ± 0.087	0.380 – 0.635	9
Sodium (%)	0.316 ± 0.033	0.258 – 0.371	11
Magnesium (%)	0.166 ± 0.010	0.148 – 0.181	11
Sulfur (%)	0.272 ± 0.059	0.208 – 0.420	10
Iron (ppm)	350.5 ± 87.3	255.0 – 523.0	11
Manganese (ppm)	92.48 ± 5.14	81.7 – 99.4	11
Zinc (ppm)	59.33 ± 10.2	46.1 – 81.6	11
Copper (ppm)	11.81 ± 2.50	8.09 – 15.4	11
Iodine (ppm)	3.54 ± 1.19	1.52 – 5.83	10
Chromium (ppm)	1.66 ± 0.46	0.85 – 2.09	11
Cobalt (ppm)	0.76 ± 0.23	0.49 – 1.15	7

TABLE I4
Contaminant Levels in NIH-07 Rat and Mouse Ration[a]

	Mean ± Standard Deviation[b]	Range	Number of Samples
Contaminants			
Arsenic (ppm)	0.40 ± 0.18	0.10 – 0.80	27
Cadmium (ppm)	0.10 ± 0.07	0.05 – 0.20	27
Lead (ppm)	0.27 ± 0.21	0.10 – 1.10	27
Mercury (ppm)	0.02 ± 0.01	0.02 – 0.50	27
Selenium (ppm)[c]	0.33 ± 0.10	0.10 – 0.44	26
Aflatoxins (ppb)[d]	<5.0		26
Nitrate nitrogen (ppm)[e]	10.77 ± 4.92	1.80 – 20.0	27
Nitrite nitrogen (ppm)[e]	0.22 ± 0.16	0.10 – 0.60	27
BHA (ppm)[f]	1.42 ± 0.90	1.00 – 4.00	26
BHT (ppm)[f]	1.31 ± 1.19	1.00 – 7.00	26
Aerobic plate count (CFU/g)	109,767 ± 105,017	4,700 – 380,000	27
Coliform (MPN/g)	17.7 ± 20.5	3.00 – 93.00	27
Escherichia coli (MPN/g)	3.3 ± 1.2	3.0 – 9.0	27
Salmonella (MPN/g)	Negative		27
Total nitrosoamines (ppb)[g]	7.00 ± 2.10	3.90 – 13.70	27
N-Nitrosodimethylamine (ppb)[g]	5.28 ± 1.45	2.90 – 9.40	27
N-Nitrosopyrrolidine (ppb)[g]	1.72 ± 1.01	1.00 – 4.70	27
Pesticides (ppm)			
α-BHC	<0.01		27
β-BHC	<0.02		27
γ-BHC	<0.01		27
δ-BHC	<0.01		27
Heptachlor	<0.01		27
Aldrin	<0.01		27
Heptachlor epoxide	<0.01		27
DDE	<0.01		27
DDD	<0.01		27
DDT	<0.01		27
HCB	<0.01		27
Mirex	<0.01		27
Methoxychlor	<0.05		27
Dieldrin	<0.01		27
Endrin	<0.01		27
Telodrin	<0.01		27
Chlordane	<0.05		27
Toxaphene	<0.10		27
Estimated PCBs	<0.20		27
Ronnel	<0.01		27
Ethion	<0.02		27
Trithion	<0.05		27
Diazinon	<0.10		27
Methyl parathion	<0.02		27
Ethyl parathion	<0.02		27
Malathion	0.27 ± 0.21	0.05 – 0.84	27
Endosulfan I	<0.01		27
Endosulfan II	<0.01		27
Endosulfan sulfate	<0.03		27

[a] CFU=colony forming units. MPN=most probable number. BHC=hexachlorocyclohexane or benzene hexachloride.
[b] For values less than the limit of detection, the detection limit is given as the mean.
[c] No selenium measurement was recorded for the lot milled 5 May 1990.
[d] No aflatoxin measurement was recorded for the lot milled 2 October 1989.
[e] Sources of contamination: alfalfa, grains, and fish meal.
[f] Sources of contamination: soy oil and fish meal. No BHA or BHT measurements were recorded for the lot milled 1 November 1989.
[g] All values were corrected for percent recovery.

APPENDIX J
SENTINEL ANIMAL PROGRAM

METHODS 184
RESULTS 185

SENTINEL ANIMAL PROGRAM

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 study 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 serology on sera from extra (sentinel) animals in the study rooms. These animals and the study animals are 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.

Serum samples were collected from randomly selected rats during the reproductive toxicity and 2-year studies. Blood from each animal was collected and allowed to clot, and the serum was separated. The samples were processed appropriately and sent to Microbiological Associates, Inc. (Bethesda, MD), for determination of antibody titers. The laboratory serology methods and viral agents for which testing was performed are tabulated below; the times at which blood was collected during the studies are also listed.

Method and Test	Time of Analysis
Reproductive Toxicity Study	
ELISA	
PVM (pneumonia virus of mice)	Study termination
RCV/SDA (rat coronavirus/sialodacryoadenitis virus)	Study termination
Sendai	Study termination
Immunofluorescence Assay	
RCV/SDA	Study termination
Hemagglutination Inhibition	
H-1 (Toolan's H-1 virus)	Study termination
KRV (Kilham rat virus)	Study termination
2-Year Study	
ELISA	
Mycoplasma arthritidis	Study termination
Mycoplasma pulmonis	Study termination
PVM	6, 12, and 18 months, study termination
RCV/SDA	6, 12, and 18 months, study termination
Sendai	6, 12, and 18 months, study termination
Immunofluorescence Assay	
RCV/SDA	12 Months
Hemagglutination Inhibition	
H-1	6, 12, and 18 months, study termination
KRV	6, 12, and 18 months, study termination

RESULTS

For the reproductive toxicity study in rats, all serology test results were negative. Two female rats had positive titers to *M. arthritidis* at the end of the 2-year study.

Further evaluation of samples positive for *M. arthritidis* by immunoblot and Western blot procedures indicated that the positive titers may have been due to a cross reaction with antibodies of nonpathogenic *Mycoplasma* or other agents. Only sporadic samples were positive, and there were no clinical findings or histopathologic changes of *M. arthritidis* infection in rats with positive titers. Accordingly, sporadic *M. arthritidis*-positive titers were considered to be false positives.