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No. 458

# TOXICOLOGY AND CARCINOGENESIS STUDIES OF BUTYL BENZYL PHTHALATE (CAS NO. 85-68-7) IN F344/N RATS (FEED STUDIES)

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.

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NTP TECHNICAL REPORT

ON THE

# TOXICOLOGY AND CARCINOGENESIS

# STUDIES OF BUTYL BENZYL PHTHALATE

(CAS NO. 85-68-7)

# IN F344/N RATS

(FEED STUDIES)

NATIONAL TOXICOLOGY PROGRAM
P.O. Box 12233
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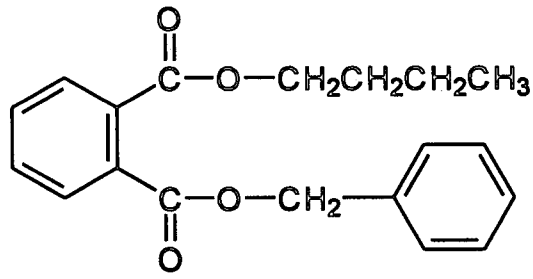
# ABSTRACT



BUTYL BENZYL PHTHALATE

CAS No. 85-68-7

Chemical Formula: $C_{19}H_{20}O_4$ Molecular Weight: 312.39

**Synonyms:** A13-14777; BBP; 1,2-benzenedicarboxylic acid butyl phenylmethyl ester (9CI); benzyl *n*-butyl phthalate; *n*-butyl benzyl phthalate; butyl phenylmethyl 1,2-benzenedicarboxylate; phthalic acid benzyl butyl ester (8CI)
**Trade names:** Palatinol BB; Santicizer 160; Sicol 160; Unimoll BB

Butyl benzyl phthalate is a plasticizer added to polymers to give flexibility and softness. It is used extensively in polyvinyl chloride and in cellulose plastics, polyvinyl acetate, polysulfides, and polyurethane. Butyl benzyl phthalate was nominated as part of a class study of phthalates. Previous studies of butyl benzyl phthalate by the NTP (1982a) resulted in chemical-related mortality in male rats beginning at about 14 weeks of exposure and, thus, were inadequate for evaluating carcinogenicity in male rats. The companion studies revealed a marginal increase in leukemia in female rats and no evidence of carcinogenicity in B6C3F$_1$ mice. Consequently, the present evaluations were conducted only in F344/N rats. Male and female F344/N rats were given butyl benzyl phthalate (at least 97% pure) in feed for 10 weeks, 26 weeks, or 2 years. Genetic toxicology studies were conducted in *Salmonella typhimurium*, L5178Y mouse lymphoma cells, cultured Chinese hamster ovary cells, mouse bone marrow cells, and *Drosophila melanogaster*.

## 10-Week Modified Mating Study in Rats

Groups of 15 male F344/N rats were given 0, 300, 2,800, or 25,000 ppm butyl benzyl phthalate (equivalent to average daily doses of approximately 20, 200, or 2,200 mg butyl benzyl phthalate/kg body weight) in feed for 10 weeks. All rats survived to the end of the study. The final mean body weight and body weight gain of the 25,000 ppm group were significantly less than those of the controls. Feed consumption by the 25,000 ppm group was less than that by the controls at the end of the study. A few minimal hematology changes occurred in the 25,000 ppm male rats. There was some evidence of a minimal anemia characterized by a decreased erythrocyte count and increases in mean cell hemoglobin and platelet count. The absolute and relative prostate gland weights of the 25,000 ppm males were significantly less than those of the controls. Degeneration of the seminiferous

tubule germinal epithelium was observed in all males from the 25,000 ppm group. The absolute right cauda, right epididymis, and right testis weights of the 25,000 ppm males were significantly less than those of the controls. The epididymal spermatozoal concentrations in 2,800 and 25,000 ppm males were significantly less than that in the controls. Although 10 females mated to 25,000 ppm males were initially found to be sperm positive, none of these females were pregnant at necropsy. The fertility indices of males and females in the 25,000 ppm group were significantly lower than those of the controls. The maternal body weights of females mated to 300 and 2,800 ppm males were similar to those of females mated to control males. There were no significant differences in litter data between the controls and the 300 and 2,800 ppm groups.

## 26-Week Study in Rats

Groups of 15 male F344/N rats were given 0, 300, 900, 2,800, 8,300, or 25,000 ppm butyl benzyl phthalate in feed for 26 weeks. Dietary levels of 300, 900, 2,800, and 8,300 ppm delivered average daily doses of approximately 30, 60, 180, and 550 mg butyl benzyl phthalate/kg body weight. The final mean body weight and body weight gain of the 25,000 ppm males were significantly less than those of the controls. Except for the 25,000 ppm males, feed consumption by all exposed groups was similar to that by the controls. An exposure-related macrocytic responsive anemia was present in the 25,000 ppm group at all time points. Additionally, minimal erythrocyte count decreases occurred sporadically in the 2,800 and 8,300 ppm groups at various time points. Reticulocyte counts were increased on days 60 and 90. Increases in mean cell hemoglobin and mean cell hemoglobin concentrations occurred in the 8,300 and 25,000 ppm rats. The absolute right cauda, right epididymis, and right testis weights and the sperm concentration of 25,000 ppm males were significantly less than those of the controls. The incidences of hypospermia and of atrophy of the seminiferous tubule in the testis and of hypospermia in the epididymis in 25,000 ppm males were significantly greater than those in the controls. Degenerative changes of the testis and epididymis in the 25,000 ppm males were qualitatively and quantitatively similar to those observed in males in the 10-week modified mating study.

## 2-Year Study in Rats

Groups of 60 male F344/N rats were given 0, 3,000, 6,000, or 12,000 ppm butyl benzyl phthalate (equivalent to average daily doses of approximately 120, 240, or 500 mg butyl benzyl phthalate/kg body weight), and groups of 60 female F344/N rats were given 0, 6,000, 12,000, or 24,000 ppm butyl benzyl phthalate (equivalent to average daily doses of approximately 300, 600, or 1,200 mg/kg) in feed for 2 years.

### *Survival, Body Weights, and Feed Consumption*

Survival of all exposed groups of male and female rats was similar to that of the controls. Mean body weights of the 12,000 ppm males and 24,000 ppm females were less than those of the controls throughout most of the study. Feed consumption by the females exposed to 24,000 ppm was less than that by the controls at the beginning of the study, but was similar to that by the controls by week 6.

### *Hematology and Hormone Assays*

In general, hematology changes were sporadic and minor. At 6 months, a minimal decrease in erythrocyte count and an increase in mean cell hemoglobin, similar to that which occurred in the 26-week study, occurred in male rats in the 12,000 ppm group. In female rats, a decreased hematocrit value occurred at 15 months in the 24,000 ppm group. There was also a mild decrease in triiodothyronine concentrations in the 24,000 ppm females at 6 and 15 months and at the end of the study.

### *Pathology Findings*

At 2 years, the incidences of pancreatic acinar cell adenoma and adenoma or carcinoma (combined) in 12,000 ppm males were significantly greater than those in the controls. The incidences of adenoma and of adenoma or carcinoma (combined) in 12,000 ppm males exceeded the ranges of historical controls from NTP 2-year feed studies. One carcinoma was observed in one 12,000 ppm male, and two adenomas were observed in 24,000 ppm females. At 2 years,

the incidence of focal hyperplasia of the pancreatic acinar cell in 12,000 ppm males was significantly greater than that in the controls.

At 2 years, transitional epithelial papillomas in the urinary bladder were observed in one control female and in two 24,000 ppm females. The incidence of this neoplasm exceeded the range of historical controls from NTP 2-year feed studies. The incidence of transitional epithelial hyperplasia in 24,000 ppm females was significantly greater than that in the controls.

The absolute right kidney weight of 12,000 ppm females and the relative right kidney weights of all exposed groups of males and of 24,000 ppm females were significantly greater than those of the controls at the 15-month interim evaluation. The severities of renal tubule pigmentation in 12,000 ppm males and in 24,000 ppm females were greater than those in the controls at 15 months and 2 years. At 2 years, the incidences of kidney mineralization in 6,000 and 24,000 ppm females were significantly less than that in the controls, and the severity was decreased in exposed females. The incidence of preputial gland adenoma or carcinoma (combined) in 12,000 ppm male rats was significantly less than in the controls, and the incidences occurred with a negative trend.

## Genetic Toxicology

Results from *in vitro* mutagenicity tests with butyl benzyl phthalate were uniformly negative. No mutagenic response was obtained in any of several strains of *Salmonella typhimurium* treated with up to 11,550 μg/plate butyl benzyl phthalate, with or without S9 metabolic activation enzymes. Negative results were also obtained in *in vitro* studies of mammalian cell systems with and without S9. No induction of trifluorothymidine resistance in L5178Y mouse lymphoma cells or sister chromatid exchanges and chromosomal aberrations in cultured Chinese hamster ovary cells were observed. These assays also were conducted with and without S9.

No significant increase in sex-linked recessive lethal mutations was observed in germ cells of male *Drosophila melanogaster* after administration of butyl benzyl phthalate either in feed or by injection.

In contrast to the negative results obtained *in vitro* and in *Drosophila*, butyl benzyl phthalate gave positive responses in two *in vivo* studies with mice. Results of a mouse bone marrow sister chromatid exchange test were positive at sample times of 23 and 42 hours, but no confirmatory test was conducted. Chromosomal aberrations were induced in bone marrow cells of male mice sampled 17 hours after intraperitoneal injection of 5,000 mg/kg butyl benzyl phthalate.

## Conclusions

Under the conditions of this 2-year feed study, there was *some evidence of carcinogenic activity** of butyl benzyl phthalate in male F344/N rats based on the increased incidences of pancreatic acinar cell adenoma and of acinar cell adenoma or carcinoma (combined). There was *equivocal evidence of carcinogenic activity* of butyl benzyl phthalate in female 344/N rats based on the marginally increased incidences of pancreatic acinar cell adenoma and of transitional epithelial papilloma of the urinary bladder.

Exposure of rats to butyl benzyl phthalate in feed for 2 years resulted in focal hyperplasia in the pancreas in male rats and in transitional epithelial hyperplasia in the urinary bladder of female rats.

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* 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 Butyl Benzyl Phthalate

| | Male F344/N Rats | Female F344/N Rats |
|---|---|---|
| **Doses** | 0, 3,000, 6,000, or 12,000 ppm in feed | 0, 6,000, 12,000, or 24,000 ppm in feed |
| **Body weights** | 12,000 ppm group less than control group | 24,000 ppm group less than control group |
| **2-Year survival rates** | 28/50, 20/50, 22/50, 22/50 | 25/50, 29/50, 29/50, 29/50 |
| **Nonneoplastic effects** | Pancreas: focal hyperplasia (4/50, 7/49, 9/50, 12/50) | Urinary bladder: transitional epithelial hyperplasia (4/50, 0/50, 1/50, 10/50) |
| **Neoplastic effects** | Pancreas: acinar cell adenoma (3/50, 2/49, 3/50, 10/50); acinar cell adenoma or carcinoma (3/50, 2/49, 3/50, 11/50) | None |
| **Uncertain findings** | None | Pancreas: acinar cell adenoma (0/50, 0/50, 0/50, 2/50)<br>Urinary bladder: transitional epithelial papilloma (1/50, 0/50, 0/50, 2/50) |
| **Level of evidence of carcinogenic activity** | Some evidence | Equivocal evidence |
| **Genetic toxicology** | | |
| *Salmonella typhimurium* gene mutations: | | Negative in strains TA98, TA100, TA1535, and TA1537 with and without S9 |
| L5178Y Mouse lymphoma gene mutations: | | No induction of trifluorothymidine resistance |
| Sister chromatid exchanges | | |
| Cultured Chinese hamster ovary cells *in vitro*: | | Negative with and without S9 |
| Mouse bone marrow *in vivo*: | | Weakly positive at 23 and 42 hours |
| Chromosomal aberrations | | |
| Cultured Chinese hamster ovary cells *in vitro*: | | Negative with and without S9 |
| Mouse bone marrow *in vivo*: | | Positive at 17 hours; negative at 36 hours |
| Sex-linked recessive lethal mutations | | |
| *Drosophila melanogaster*: | | No induction of sex-linked recessive lethal mutations |

# 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 butyl benzyl phthalate on 20 June 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.

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## SUMMARY OF TECHNICAL REPORTS REVIEW SUBCOMMITTEE COMMENTS

On 20 June 1995, the draft Technical Report on the toxicology and carcinogenesis studies of butyl benzyl phthalate 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. F.W. Kari, NIEHS, introduced the toxicology and carcinogenesis studies of butyl benzyl phthalate by describing the uses of the chemical and the rationale for study, describing the experimental design, reporting on survival and body weight effects, and commenting on chemical-related neoplastic lesions in male rats and uncertain neoplastic findings in female rats as well as chemical-related nonneoplastic lesions in male and female rats. Dr. Kari noted that there had been an earlier 2-year NTP study with butyl benzyl phthalate in which there was no evidence of adverse effects in mice and a marginally increased incidence of mononuclear cell leukemia in female rats. This study was inadequate in male rats due to high mortality beginning around week 14. Thus, the design of the subchronic phase of the current study in male rats was more elaborate in an attempt to determine if early mortality would be a problem. The design included a 10-week modified mating trial as well as other indices of reproductive toxicity. The proposed conclusions for the 2-year study were *some evidence of carcinogenic activity* in male F344/N rats and *equivocal evidence of carcinogenic activity* in female F344/N rats.

Dr. Reddy, a principal reviewer, agreed with the proposed conclusions although he thought that the proposed conclusions from the previous 2-year study in mice should be cited. Dr. Kari responded that this information would be added to the Abstract. Dr. Reddy noted the 20% incidence of pancreatic acinar cell adenoma versus only a 2% incidence of carcinoma in 12,000 ppm male rats. He asked for definition of the criteria for distinguishing adenomas from carcinomas since, based on his experience, he wondered if carcinomas were underrepresented. Dr. J.R. Hailey, NIEHS, said more would be added concerning how the distinction was made between adenomas and carcinomas (p. 43). While metastasis was an easy marker for a carcinoma, other features were used such as cellular pleomorphism or tremendous heterogeneity in the growth pattern, cellular atypia, and high mitotic index.

Dr. Klaassen, the second principal reviewer, agreed with the proposed conclusions. He said that reference to the mouse studies should be made earlier in the Technical Report, preferably in the Abstract, and that more information should be included on phthalate carcinogenicity in regard to the rationale (p. 20). Because of the known effects of phthalates on male reproduction, Dr. Klaassen was pleased to see reproduction studies in this Technical Report.

Dr. Ryan, the third principal reviewer, agreed with the proposed conclusions. She wondered whether there should also be a conclusion regarding the reproductive toxicity. Dr. Ryan found it worrisome that so many animals died in the 26-week study during anesthesia prior to blood sampling and asked whether there was any bias here whereby weaker or sicker animals were more likely to die during the procedure. Dr. Kari said he could only speculate that the higher ratio of carbon dioxide to oxygen than generally used may have contributed to the excessive mortality. It did not appear to be due to chemical interaction, because the numbers of deaths in control animals were similar to those in exposed groups of animals.

Dr. Miller noted the National Institute for Occupational Safety and Health (NIOSH) reference that over 300,000 workers were potentially exposed to butyl benzyl phthalate and asked whether there were data about occupational exposure in terms of airborne concentrations. Dr. J. Haartz, NIOSH, commented that their database does not have such quantitative information. She reported that the Occupational Safety and Health Administration does not have an exposure database and offered to follow up on that for Dr. Kari.

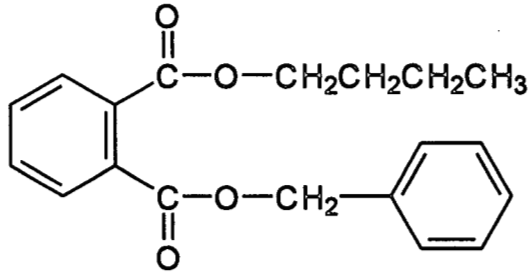
Dr. M. Stevens, Manager of Toxicology Projects, Monsanto Business Services, stated that the biggest

concern of the Monsanto Company, a primary maker of butyl benzyl phthalate, was that this study not be considered in isolation from theirs and studies of others. He pointed out that survival and neoplasm findings in the earlier NTP study were not repeated in the current study, and, further, when diet restriction was employed, the increased incidences of pancreatic neoplasms seen in the current study were eliminated. Thus, looking at the multiple studies with no consistent reported findings, Dr. Stevens thought a decision could not be made about the potential carcinogenicity of butyl benzyl phthalate.

Dr. Miller asked the industry representatives for information on occupational and consumer exposures. Dr. R. Hogue, Monsanto Company, said occupational exposure is quite low, as is consumer exposure in vinyl flooring, because the butyl benzyl phthalate is bound into a polymeric system. He said exposure data would be provided to the NTP. Dr. R.W. Hart, NCTR, returned to the effects of feed restriction on neoplasm incidence, noting that in 30-month feed restriction male rats, three exposed rats developed pancreatic acinar cell adenomas. In feed-restricted exposed females at 30 months, there was a statistically significant increased incidence of neoplasms of the urinary bladder.

Dr. Ryan moved that the Technical Report on butyl benzyl phthalate be accepted with the revisions discussed and with the conclusions as written for male rats, *some evidence of carcinogenic activity*, and for female rats, *equivocal evidence of carcinogenic activity*. Dr. Reddy seconded the motion, which was accepted unanimously with 10 votes.

# INTRODUCTION



BUTYL BENZYL PHTHALATE

CAS No. 85-68-7

Chemical Formula: $C_{19}H_{20}O_4$ Molecular Weight: 312.39

**Synonyms:** A13-14777; BBP; 1,2-benzenedicarboxylic acid butyl phenylmethyl ester (9CI); benzyl *n*-butyl phthalate; *n*-butyl benzyl phthalate; butyl phenylmethyl 1,2-benzenedicarboxylate; phthalic acid benzyl butyl ester (8CI)
**Trade names:** Palatinol BB; Santicizer 160; Sicol 160; Unimoll BB

## CHEMICAL AND PHYSICAL PROPERTIES

Butyl benzyl phthalate is a clear, oily liquid with a slight odor and a bitter taste. It has a melting point of -35° C, a boiling point of 370° C, and a specific gravity of 1.116 at 25° C. (Sax and Lewis, 1989). Phthalate acid esters generally have a low volatility at standard temperature and pressure, particularly those of the long-chain and branched compounds such as butyl benzyl phthalate and di(2-ethylhexyl)phthalate (Kluwe, 1982). Butyl benzyl phthalate has a vapor pressure of $8.6 \times 10^{-6}$ mm Hg at 20° C and a vapor density of 10.8. It is combustible when exposed to heat or flame, with a flash point of 199° C and an autoignition temperature of 233° C; when heated to decomposition, it emits acrid smoke and irritating fumes. Butyl benzyl phthalate can react with oxidizers, and it is soluble in most organic solvents and in dimethylsulfoxide, acetone, and 95% ethanol (less than 100 mg/mL at 23° C) (Verschueren, 1983; *Hawley's*, 1987; Sax and Lewis, 1989).

Phthalate esters with short alkyl groups are appreciably soluble in water. However, most other dialkyl phthalates are relatively insoluble in aqueous media because of their lipophilic structures; butyl benzyl phthalate is almost insoluble in distilled water (2.69 mg/L at 20° C; Howard *et al.*, 1985) and in deionized well water (2.9 mg/L at 20° C; Gledhill *et al.*, 1980). The lipophilicity of butyl benzyl phthalate is also suggested by its octanol/water partition coefficient (log $K_{ow}$), which has been reported as 4.91 (Leyder and Boulanger, 1983) and 4.77 (Gledhill *et al.*, 1980).

## PRODUCTION, USE, AND HUMAN EXPOSURE

Phthalate acid esters are synthesized commercially by condensation of an appropriate alcohol with phthalic anhydride in the presence of an acid catalyst (Kluwe, 1982). Butyl benzyl phthalate has been commercially

produced in the United States since 1946 (U.S. Tariff Commission, 1948). The annual production of butyl benzyl phthalate in the United States was estimated as 80 million pounds per year (USITC, 1978).

Butyl benzyl phthalate is a plasticizer added to polymers (generally at concentrations of 50 to 75 parts per 100 parts of resin) to provide flexibility and softness. It is used extensively in polyvinyl chloride for vinyl floor tile, vinyl foams, carpet backings, and Astroturf®. Other polymers plasticized with butyl benzyl phthalate include cellulose plastics, polyvinyl acetate, polysulfides, and polyurethane. Butyl benzyl phthalate is an organic intermediate and a component of synthetic leather, biodegradable tampon applicators, automotive paint and upholstery, acrylic caulks, plastic filler for automobile body repair, air conditioning filters, inks, and tacky adhesives used to secure medical devices such as ostomy bags and compresses to the skin. It is also used as a carrier and dispersant for pesticides, colorants, solvents, catalysts, munitions, industrial oils, insect repellents, and perfumes and as a plasticizer in aerosol hairsprays at concentrations of less than 1% (Statsek, 1974; Singmaster and Crosby, 1976; Lawrence, 1979; USEPA, 1981; Eigenberg *et al.*, 1986; CIRP, 1992).

The U.S. Food and Drug Administration (FDA) has approved butyl benzyl phthalate, provided that it contains no more than 1% by weight of dibenzylphthalate, for use in packaging and wrapping materials and polyvinyl acetate emulsion adhesives in contact with food products (21 CFR, §§ 175.105, 175.300, 175.320, 176.170, 176.180, 177.2420, 178.3740). Regenerated cellulose film is coated with plasticized nitrocellulose containing butyl benzyl phthalate to provide flexibility and heat sealability; it is used primarily to wrap confectionery products, pastries, cakes, and sandwiches. Studies have shown that the flexibility of these films results from dispersion of the various phthalate plasticizers in the matrix of the polymer chains, which decreases the interactive forces of adjacent chains (Autian, 1973). However, the lack of chemical bonding with the polymer allows plasticizers to migrate into foods at a rate of less than 0.05% to 1.8% (less than 0.05% to 0.8% for butyl benzyl phthalate) on a total film-weight basis from nitrocellulose film containing 30% to 40% plasticizer. Food products have been shown to contain 0.5 to 53 mg butyl benzyl phthalate/kg, individually or in combination with other plasticizers (Castle *et al.*, 1988).

Retail samples of Canadian butter and margarine wrapped in aluminum foil-paper laminate were found to contain butyl benzyl phthalate, dibutylphthalate, and/or di(2-ethylhexyl)phthalate at concentrations up to 47.8, 10.6, and 11.9 $\mu$g/g, respectively. Analyses of the food wrapping materials indicated that these phthalate esters migrated from the protective washcoat on the exterior surface of the wrappers and through the food-contacting surface into the food (Page and Lacroix, 1992). Migration into food of plasticizers used in ink on the outer surface of commercially printed polypropylene films has also been demonstrated; this migration increases with storage time of the wrapped product (Castle *et al.*, 1989). Concern regarding the potential toxicity of phthalate esters as contaminants of food led the Japanese Ministry of Health and Welfare to establish regulations to limit the migration of these compounds into foodstuffs and beverages (Omori, 1976).

The U.S. Environmental Protection Agency (EPA) has exempted butyl benzyl phthalate from tolerance requirements when used as an inert ingredient in laminated dispensers for controlled release of Glossypure®, a synthetic pheromone used on cotton to disrupt the mating of the pink bollworm (USEPA, 1981).

Occupational exposure to butyl benzyl phthalate was examined in a National Occupational Hazard Survey conducted by the National Institute of Occupational Safety and Health (NIOSH) covering the years from 1972 to 1974. During this time, an estimated 68,488 workers were potentially exposed to butyl benzyl phthalate (NIOSH, 1976). In a second survey of workplace exposure (National Occupational Exposure Survey) covering the years from 1981 to 1983, an estimated 331,840 workers were potentially exposed to butyl benzyl phthalate (NIOSH, 1990). However, there is no Occupational Safety and Health Administration permissible exposure limit for butyl benzyl phthalate, nor is there an American Conference of Governmental Industrial Hygienists recommended threshold limit value-time weighted average reported in the current literature.

Butyl benzyl phthalate can enter the environment as a consequence of its manufacture, use, and disposal. Humans may be exposed to butyl benzyl phthalate in occupational surroundings and from environmental contamination. Based on use patterns for vinyl flooring and food packaging materials including adhesives, almost all consumers are exposed to butyl benzyl phthalate to some extent.

## Regulatory Status

Butyl benzyl phthalate is classified by the FDA as a migratory, indirect food additive (21 CFR, § 170.3). It has been designated by the EPA as a base-neutral priority pollutant as well as a hazardous waste to be disposed of according to the federal hazardous waste management system (40 CFR, § 261.33). Butyl benzyl phthalate is a regulated substance under the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA). Under CERCLA, releases of butyl benzyl phthalate into the environment in quantities exceeding 100 pounds must be reported to the EPA (40 CFR, Part 302).

## Environmental Impact

STORET, the EPA water quality database, was used to determine the concentrations of butyl benzyl phthalate throughout the United States in all types of water samples (mean aqueous concentration of 9 μg/L), sediment (median concentration of less than 500 μg/kg dry, 6% detectable), and biota (median concentration of less than 2.5 μg/kg wet, 3% detectable) (Staples *et al.*, 1985). In studies by several groups of investigators, butyl benzyl phthalate was identified in drinking water in New Orleans (0.08 to 1.8 μg/L) and Philadelphia (0.3 to 1 μg/L), surface water in Illinois, Michigan, Missouri, New Jersey, and Pennsylvania (0.2 to 2.4 μg/L), and industrial wastewater effluents near Philadelphia (40 μg/L). These effluents and others were treated at a local wastewater treatment plant which discharged effluents containing 100 μg butyl benzyl phthalate/L into the Delaware river (Keith *et al.*, 1976; Shackelford and Keith, 1976; Sheldon and Hites, 1978, 1979). Sediment samples collected from the Saginaw River in Michigan and the Missouri River at Weldon Springs, Missouri, contained butyl benzyl phthalate at concentrations of 400 to 567 ng/g and 100 ng/g, respectively (Gledhill *et al.*, 1980). Butyl benzyl phthalate was identified at a concentration of 0.8 ng/g in sediment from Lake Pontchartrain in New Orleans (McFall *et al.*, 1985).

Studies performed in water treatment plants have demonstrated that biodegradation of phthalates is temperature dependent and can occur under aerobic or anaerobic conditions. A wide range of organisms in soil, river and sea water, sediments, and water treatment plants are capable of biodegrading phthalates. Longer alkyl chain phthalates biodegrade more slowly than do those with shorter chains. Elimination from natural waters could be by biodegradation, photodegradation, volatilization, or adsorption onto sediments; phthalate concentrations in sediments are markedly higher than those in the water above the sediment strata. In the opinion of the ECETOC Task Force, the widespread disposition of low concentrations of phthalate esters in surface water may reflect a balance between continuous input to the aquatic environment and elimination of the compounds by biodegradation (ECETOC, 1985).

## Absorption, Distribution, Metabolism, and Excretion

### *Experimental Animals*

Phthalic acid esters are generally well absorbed from the gastrointestinal tract. Due to the lipophilic nature of these compounds, absorption also occurs via dermal and pulmonary routes (Kluwe, 1982).

Both ester linkages of phthalate esters can be hydrolyzed, leaving phthalic acid as a product. Esterases that rapidly hydrolyze a single ester group to generate the monoester metabolite are present in several mammalian tissues, including intestinal mucosal cells, and extracellular enzymes present in the intestinal contents are also capable of hydrolyzing phthalate esters (Rowland *et al.*, 1977). Butyl benzyl phthalate is partially hydrolyzed by intestinal esterases, primarily to monobutylphthalate and benzyl alcohol with monobenzylphthalate and *n*-butanol as minor products of hydrolysis (Agarwal *et al.*, 1985).

Tissue distribution and pharmacokinetic studies by Eigenberg *et al.* (1986) demonstrated that butyl benzyl phthalate is rapidly cleared from the body of male F344 rats administered gavage (2 to 2,000 mg/kg) or

intravenous (20 mg/kg) doses of [$^{14}$C]-butyl benzyl phthalate. Excretion of 75% to 86% of a gavage dose occurred within 24 hours, and greater than 92% was eliminated by the end of 4 days. The major route of elimination of butyl benzyl phthalate in rats is excretion of metabolites into the bile. At gavage doses from 2 to 200 mg/kg, extensive enterohepatic reabsorption of these metabolites or their degradation products resulted in urinary excretion of 75% of the dose; another 20% was eliminated in the feces. At 2,000 mg/kg, however, 72% of the dose was eliminated in the feces, with only 22% excreted in the urine. This shift from urinary to fecal elimination may be due to incomplete absorption of butyl benzyl phthalate or its metabolites during enterohepatic recirculation. The half-lives of butyl benzyl phthalate and its metabolites in all tissues are approximately 6 hours; although lipophilic, butyl benzyl phthalate is not sequestered in fat due to its rapid metabolism.

Pharmacokinetic studies in four beagle dogs administered a divided oral dose equivalent to 5 g butyl benzyl phthalate/kg body weight over a period of 4 hours demonstrated recovery of 88% (male) and 91% (female) of unchanged butyl benzyl phthalate from the feces; approximately 4% was excreted as urinary phthalic acid (Erickson, 1965).

The major urinary metabolites of butyl benzyl phthalate in rats are monophthalate derivatives (10% to 40% of the dose) and monophthalate glucuronides (2% to 21%) (Eigenberg *et al.*, 1986). Since butyl benzyl phthalate is an asymmetric diester, two different monophthalates are produced, monobutylphthalate or monobenzylphthalate. There is a preference for hydrolysis of the benzyl ester, resulting in a preponderance (approximately 3:1) of monobutylphthalate in the urine. The urinary half esters of other phthalates, such as diethylphthalate and dibutylphthalate, have been shown to be four times more toxic than their parent compounds (International Labor Office, 1983).

Dermal absorption of [$^{14}$C]-radiolabeled butyl benzyl phthalate and other phthalate esters was evaluated in male F344 rats using urinary and fecal excretion as an index of relative percutaneous absorption (Elsisi *et al.*, 1989). By day 7, 30% of a 157 $\mu$mol butyl benzyl phthalate/kg dose was excreted in the urine and feces, while 44.9% and 6.3% remained in the skin of the application site and in the occlusive plastic cap over the site, respectively. Five percent of the applied dose was retained in the tissues after 7 days, 4.6% in muscle and 0.5% in the brain, spinal cord, and testes.

Recovery of radioactivity from fetal tissues following treatment of maternal Sprague-Dawley rats with [$^{14}$C]-di(2-ethylhexyl)phthalate or [$^{14}$C]-diethylphthalate demonstrated that maternal-fetal transfer of dialkyl phthalate esters occurs across the placenta (Singh *et al.*, 1975). Since lipophilic chemicals readily partition into high fat materials such as breast milk, excretion of phthalates into maternal milk is also probable.

### *Humans*

The most common route of exposure to phthalate esters in humans is ingestion of food or liquids (Kluwe, 1982). However, no information related to the pharmacokinetics and metabolism of butyl benzyl phthalate in humans has been reported in the literature.

## TOXICITY

### *Experimental Animals*

Phthalate acid esters have very low acute toxicity by oral and parenteral routes; the acceptable daily oral intake values for phthalates are of the same order of magnitude as some chemicals approved for use as direct food additives (Krauskopf, 1973). Absorption through the skin occurs to some extent, with $LD_{50}$ values in rabbits ranging from 3.4 mL/kg for diallylphthalate to 20 mL/kg for dibutylphthalate and dihexylphthalate (*Patty's*, 1967). However, due to their low volatility, inhalation of most phthalates is not associated with acute toxic effects (Autian, 1973).

The acute lethal toxicity of phthalate esters, administered orally or parenterally, is inversely proportional to molecular weight; esters with the shorter alkyl groups are the most water soluble and exhibit the greatest toxicity (Autian, 1973). Butyl benzyl phthalate has a relatively high molecular weight and is almost insoluble in water. The gavage $LD_{50}$ values for butyl benzyl phthalate in a corn oil vehicle, calculated 14 days after administration of a single dose, were 2.33 g/kg for male and female Fisher 344/N rats, 6.16 g/kg for male B6C3F$_1$ mice, and 4.17 g/kg for female B6C3F$_1$ mice (NTP, 1982a). The oral $LD_{50}$ in Sprague-Dawley rats was 20.4 g/kg

(Shibko and Blumenthal, 1973; Hammond *et al.*, 1987). The intraperitoneal $LD_{50}$ of butyl benzyl phthalate in Swiss Webster mice was 3.16 g/kg (Calley *et al.*, 1966).

In a 14-day feed study of butyl benzyl phthalate in F344/N rats, exposure to 25,000 ppm or more resulted in lower body weight gains than those in the controls. Thymic atrophy occurred in all 100,000 ppm rats, and testicular degeneration was observed in all 50,000 and 100,000 ppm males. No compound-related effects were observed in a companion study in $B6C3F_1$ mice administered concentrations up to 25,000 ppm in feed. In similar 13-week feed studies, lower body weight gains and testicular degeneration, characterized by loss of the germinal epithelium of the seminiferous tubules, were observed in male rats receiving concentrations of 25,000 ppm, whereas compound-related effects in mice were limited to lower body weight gains in male mice exposed to concentrations of 1,600 ppm or more and in 12,500 and 25,000 ppm females (NTP, 1982a). The testicular degeneration observed in rats might have been related to conversion of butyl benzyl phthalate to its monobutylphthalate metabolite, which has been shown to produce testicular atrophy (Foster *et al.*, 1981; Gangolli, 1982).

Sprague-Dawley rats exposed to concentrations of 2,000 to 4,000 mg butyl benzyl phthalate/kg in feed for 2 weeks exhibited dose-related posterior body stiffness and incoordination of the hind limbs, which was more severe in males. These signs generally disappeared by the end of a 1-week recovery period (Hammond *et al.*, 1987). In a follow-up 6-week neurological study, body weight gains of Charles River CD rats exposed to 1,500 or 3,000 mg butyl benzyl phthalate/kg body weight were lower than those of the controls, and transient hind limb stiffness was observed in the 3,000 mg/kg group, predominantly in males. Histopathologic examination of tissues from the central and peripheral nervous systems of these animals failed to reveal compound-related pathologic changes (Robinson, 1991).

The no-observable-effect-level (NOEL) in a 90-day study in rats administered feed containing up to 2% butyl benzyl phthalate by weight was 0.5%. Body weight gains of rats exposed to 1.5% and 2% were slightly lower than those of the controls, and liver weights were greater than those of the controls at concentrations of 1% and above (Erickson, 1965). In a similar 90-day feed study in dogs, the NOEL of butyl benzyl phthalate was 2%. Dogs exposed to 5% butyl benzyl phthalate gained less weight initially due to refusal to eat; feed consumption returned to normal after capsule dosing was instituted on day 40 of the study (Erickson, 1965).

After 14 weeks of a 2-year study in F344/N rats exposed to 6,000 or 12,000 ppm butyl benzyl phthalate in feed, compound-related mortality in males resulted from unexplained internal hemorrhaging; all surviving male rats were sacrificed at week 29 or 30 (NTP, 1982a).

The comparative parenteral toxicity of a series of phthalate esters was evaluated by Calley *et al.* (1966). In a 6-week study, Swiss Webster mice given daily intraperitoneal injections of 500 mg butyl benzyl phthalate/kg body weight exhibited a slight retardation in weight gain compared to controls, extramedullary hematopoiesis in the liver and spleen, and periportal hepatitis in addition to an acute peritonitis caused by the daily intraperitoneal injections. As in acute studies, the relative toxicity of the phthalate esters tested was directly related to water solubility and molecular weight. In the same series of studies, the order of potentiation of hexobarbital narcosis also followed the water solubility of the compounds. A 500 mg/kg intraperitoneal injection of butyl benzyl phthalate emulsified in 3% acacia, given 30 minutes prior to a 60 mg/kg intraperitoneal injection of sodium hexobarbital, resulted in a sleep time of 62 minutes, which was significantly greater than that of the controls (46 minutes; $P>0.05$); this was interpreted by the authors to be an indication of acute depression of the central nervous system.

Intradermal irritation caused by phthalate esters in rabbits was evaluated following injection of 1 mL/kg of 1% trypan blue into the marginal ear vein. Mild to moderate inflammatory responses occurred with butyl benzyl, dibutyl, diisobutyl, di-(methoxyethyl), and dicapryl phthalates, as indicated by leakage of dye at the injection site 15 to 26 minutes after the injection, whereas dimethyl, diethyl, and di(2-ethylhexyl) phthalates elicited marked inflammatory responses, indicated by rapid and intense concentration of extravasated Trypan Blue at the injection sites. With the

exception of di(2-ethylhexyl)phthalate, the irritant activity of this series of esters was related to molecular weight (Calley *et al.*, 1966).

Although some of the most water soluble phthalate esters [dimethyl, diethyl, and di(methoxyethyl) phthalates] cause *in vitro* cytotoxicity in strain L 929 mouse fibroblasts, 0.05 mL of a 50 mg/mL emulsion of butyl benzyl phthalate was not toxic to mouse fibroblasts. None of the phthalate esters tested at the same concentration were toxic to chick embryo cells (Calley *et al.*, 1966). In another *in vitro* test, a butyl benzyl phthalate concentration of $1.8 \times 10^{-4}$ M slightly inhibited the growth of cultured neonatal rat cerebellar cells, and cellular toxicity was observed with concentrations of $7 \times 10^{-4}$ M or more (Kasuya, 1980; Teranishi and Kasuya, 1980). Butyl benzyl phthalate has been shown to be cytotoxic to HeLa cells in a 24-hour metabolic inhibition test (Ekwall *et al.*, 1982).

### *Humans*

Butyl benzyl phthalate is a slight skin, eye, and mucous membrane irritant as well as a central nervous system depressant (Gosselin *et al.*, 1984). Occupational studies have indicated that exposure to high concentrations of mixed phthalates in workplace air may produce transient irritation of the nose and throat (*Patty's*, 1967). Prolonged contact with the liquid caused irritation of the eyes and skin (CHRIS, 1992). Other symptoms include respiratory irritation, respiratory distress, lightheadedness, nausea, pharyngitis, sleepiness, and erythema (Hazardline, 1983). Repeated exposure to phthalate vapors mixed with other plasticizers may lead to neurotoxic effects such as pain, numbness, and weakness of the extremities; a higher incidence of toxic polyneuritis; and a lowering of the excitability of the vestibular and olfactory receptors (Milkov *et al.*, 1973).

## REPRODUCTIVE AND DEVELOPMENTAL TOXICITY

### *Experimental Animals*

Butyl benzyl phthalate had no adverse effect on embryo development or neonatal death rates at 21 days of age in two studies in the chick embryo system. However, compound-related lethality (as evidenced by a decrease of hatching rates) was observed in one study (Haberman *et al.*, 1968), but not in the other (Bower *et al.*, 1970). In another study, the embryotoxicity of butyl benzyl phthalate was relatively limited (Korhonen *et al.*, 1983).

In a series of feed or gavage studies, Ema *et al.* (1990, 1991, 1992a,b, 1993) administered butyl benzyl phthalate to pregnant Wistar rats throughout gestation in order to evaluate compound-related developmental deviations caused at any time during the span of development (e.g., death of the developing organism, structural abnormalities, and growth retardation) using criteria described by Wilson (1973). The NOELs for butyl benzyl phthalate administered daily in feed on gestation days 0 through 20 were 0.5% (375 mg/kg) for maternal toxicity and 1% (654 mg/kg) for embryo-fetal toxicity. Adjusted maternal body weight gain and feed consumption during pregnancy were significantly lower in the 1% and 2% (974 mg/kg) groups than those in the controls. In the 2% group, all pregnant rats exhibited complete resorption of all implanted embryos (Ema *et al.*, 1990).

A subsequent study was performed to rule out maternal toxicity (malnutrition during pregnancy) as a contributing factor in the embryolethality observed in treated rats with the addition of a feed-restricted (pair-fed) group that received approximately the same amount of feed consumed by the 2% butyl benzyl phthalate group (Ema *et al.*, 1991). As in the previous study, complete resorption of all implanted embryos occurred in the 2% butyl benzyl phthalate group but did not occur in any of the pair-fed rats, indicating that embryolethality was a direct effect of butyl benzyl phthalate and/or its metabolites.

Pregnant Wistar rats given concentrations of 0.2% butyl benzyl phthalate in feed on gestation days 0 through 20, 0 through 11, or 11 through 20 consumed less feed and gained less weight than the control and pair-fed groups. All dams exposed to butyl benzyl phthalate on days 0 through 20 or 0 through 11 exhibited complete resorption of all implanted embryos. No increase in post-implantation loss occurred in rats exposed on days 11 through 20. However, the incidence of fetal malformations in this

group was significantly higher than in the control and pair-fed groups, consisting predominantly of cleft palate (72/134 fetuses) and fusion of the sternebrae. The results of this study suggested that the susceptibility to the teratogenic effect of butyl benzyl phthalate varies with the developmental stage at the time of administration. Butyl benzyl phthalate exposure during the first half of pregnancy resulted in embryolethality; similar exposure during the second half of pregnancy caused marked teratogenicity (Ema *et al.*, 1992a). Embryolethality in experimental animals exposed to plasticizers was also reported in an earlier Russian study (Aldyreva *et al.*, 1975).

Another study by Ema *et al.* (1993) demonstrated that the susceptibility to the teratogenic effect of butyl benzyl phthalate varies with the developmental stage at the time of administration. Butyl benzyl phthalate caused a dose-related increased incidence of malformed fetuses in rats administered gavage doses of 0.6, 0.75, or 1 g/kg on gestation days 7 through 9 or 13 through 15, but was not teratogenic when given on gestation days 10 through 12. Treatment on days 7 through 9 resulted in skeletal malformations such as fusion of the cervical vertebral arches and deformity of the thoracic vertebrae, whereas treatment on days 13 through 15 resulted in external malformations and skeletal malformations such as cleft palate and fusion of the sternebrae. In a similar feed study, cleft palate occurred in 95% of the fetuses of dams exposed to 2% butyl benzyl phthalate on days 7 through 16; fusion of the sternebrae was also a common observation in this group (Ema *et al.*, 1992b).

The maternal and developmental NOELs in Sprague-Dawley (CD) rats and Swiss (CD-1®) mice exposed to butyl benzyl phthalate in feed on gestation days 6 through 15 were 0.5% (419 mg/kg per day) and 0.1% (182 mg/kg per day), respectively (Price *et al.*, 1990). In rats exposed to 1.25% (1,102 mg/kg per day) and mice exposed to 0.5% (910 mg/kg/day), dam weight gains were lower and the incidence of fetal reabsorptions and malformations were greater than the controls. The developmental NOEL in rabbits administered butyl benzyl phthalate on gestation days 6 through 18 was 10 mg/kg (Hammond, 1981).

Other phthalate acid esters that have been shown to exhibit teratogenicity in rats and/or mice include dimethoxyethyl, diethyl, dibutyl, diisobutyl, butyl carbobutoxymethyl, dioctyl, dipropyl, dicyclohexyl, di(2-ethylhexyl), and mono(2-ethylhexyl) phthalates (Singh *et al.*, 1972, 1973; Yagi *et al.*, 1980).

### *Humans*

Increased incidences of anovulatory reproductive cycles and low estrogen concentrations were reported among Russian women working with phthalate plasticizers; the abnormal cycles were associated with spontaneous abortion (Aldyreva *et al.*, 1975). However, the specific phthalates implicated, dose levels, and other data were not reported.

## CARCINOGENICITY

### *Experimental Animals*

Di(2-ethylhexyl)phthalate, a structurally related compound, was shown to be an hepatic carcinogen in a 2-year feed study in F344/N rats exposed to 6,000 or 12,000 ppm and B6C3F$_1$ mice exposed to 3,000 or 6,000 ppm (NTP, 1982b). The incidences of hepatocellular carcinoma were significantly greater than those of the controls in 12,000 ppm female rats, 6,000 ppm male mice, and 3,000 and 6,000 ppm female mice and occurred with positive trends in female rats, male mice, and female mice. In male rats, the combined incidences of hepatocellular carcinoma or neoplastic nodules were significantly greater than in the controls.

In a carcinogenesis bioassay of butyl benzyl phthalate administered to F344/N rats in feed containing 6,000 or 12,000 ppm for approximately 2 years (NTP, 1982a), the incidence of mononuclear cell leukemia, generally characterized by splenomegaly and often by hepatomegaly, was significantly greater in 12,000 ppm females than that in the controls and occurred with a positive trend. The incidence of mononuclear cell leukemia in the 12,000 ppm females was also significantly greater than that in historical controls from NTP 2-year feed studies, and the overall trend remained statistically significant. However, the results of the study were inadequate to evaluate carcinogenicity in male rats due to unexplained compound-related toxicity and early mortality beginning at 14 weeks; all surviving males were sacrificed at 29 to 30 weeks. There was no evidence of carcinogenicity of butyl benzyl phthalate in the companion study in B6C3F$_1$ mice. In the carcinogenesis bioassay of diallyphthalate, the incidence of

mononuclear cell leukemia was also significantly greater in female rats administered gavage doses of 100 mg/kg than that in the controls (NTP, 1985).

Butyl benzyl phthalate, along with other drinking water contaminants, was tested for carcinogenicity and failed to induce a positive pulmonary adenoma response in strain A male mice administered intraperitoneal doses up to 800 mg/kg in tricaprylin 3 times per week for a total of 24 injections (19,200 mg/kg) (Theiss *et al.*, 1977).

### *Humans*

According to the International Agency for Research on Cancer, no evaluation could be made to determine the carcinogenic risk of butyl benzyl phthalate to humans (IARC, 1982, 1987).

## GENETIC TOXICITY

There are few mutagenicity data published for butyl benzyl phthalate other than the data included in Appendix C of this Technical Report, and with the exception of the two unpublished *in vivo* studies discussed in Appendix C, all results were negative. Butyl benzyl phthalate did not induce gene mutations in *Salmonella typhimurium* tested with either a standard plate incorporation protocol (Kozumbo *et al.*, 1982) or modified preincubation protocol (Zeiger *et al.*, 1985; Table C1), with and without S9.

In mammalian cell systems, butyl benzyl phthalate did not induce mutations at the TK locus of L5178Y mouse lymphoma cells (Myhr and Caspary, 1991; Table C2), and it did not induce sister chromatid exchanges or chromosomal aberrations in cultured Chinese hamster ovary cells (Galloway *et al.*, 1987; Tables C3 and C4). These tests were conducted with and without Aroclor-induced S9.

No increases in sex-linked recessive lethal mutations were observed in germ cells of male *Drosophila melanogaster* treated with butyl benzyl phthalate by injection or in dosed feed (Valencia *et al.*, 1985; Table C5).

There is only one published mammalian *in vivo* study with butyl benzyl phthalate, an abstract by Bishop *et al.* (1987) describing results of a dominant lethal test in male mice. After treatment of male B6C3$F_1$ and CD-1 mice with three subcutaneous injections of butyl benzyl phthalate (doses up to 4,560 mg/kg per day) and matings to untreated virgin females of the same strain for a period of 49 days, no increase in fetal deaths and no decrease in various fertility parameters were found. Butyl benzyl phthalate was concluded to be negative in the mouse dominant lethal assay. Two unpublished *in vivo* studies showing induction of sister chromatid exchanges and chromosomal aberrations in bone marrow cells of male mice administered butyl benzyl phthalate by a single intraperitoneal injection are described in detail in Appendix C of this report.

In summary, all mutagenicity tests with butyl benzyl phthalate were negative, with the exception of two unpublished mouse *in vivo* tests demonstrating weak chromosomal effects.

## STUDY RATIONALE

Butyl benzyl phthalate was nominated by the EPA, along with several other phthalates, as part of a class study evaluation. Diethylhexyladipate and di(2-ethylhexyl)phthalate have been implicated as rodent carcinogens (NTP, 1982b,c), and diallylphthalate had equivocal evidence of carcinogenicity in female rats (NTP, 1985) and male mice (NTP, 1983a). In an earlier study, carcinogenicity of butyl benzyl phthalate in rats could not be evaluated due to an unexplained compound-related toxicity and early mortality in males (NTP, 1982a). The oral route of exposure was chosen for the current studies to mimic the principal means of human exposure to butyl benzyl phthalate. Since the reproductive morphology and function of male rats and mice have been affected by other phthalate esters, a 10-week modified mating study was conducted in rats to assess the possibility that butyl benzyl phthalate could also adversely affect male reproduction (Gray and Gangolli, 1986; Lamb *et al.*, 1987; Heindel *et al.*, 1989).

# MATERIALS AND METHODS

## PROCUREMENT AND CHARACTERIZATION OF BUTYL BENZYL PHTHALATE

Butyl benzyl phthalate was obtained from Chem Central (Kansas City, MO) in two lots (C090882 and L-121 1-87). Lot C090882 was used during the 10-week modified mating study and the 26-week study, and lot L-121 1-87 was used during the 2-year study. 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 butyl benzyl phthalate studies are on file at the National Institute of Environmental Health Sciences (NIEHS).

The chemical, a clear, colorless, viscous liquid, was identified as butyl benzyl phthalate by infrared, ultraviolet/visible, and nuclear magnetic resonance spectroscopy. The purity of both lots was determined by elemental analyses, Karl Fischer water analysis, functional group titrations (free acid titration and ester hydrolysis), thin-layer chromatography, and gas chromatography. For lot C090882, elemental analyses for carbon and hydrogen were in agreement with the theoretical values for butyl benzyl phthalate. Karl Fischer water analysis indicated 0.23% ± 0.01% water. Functional group titrations indicated free acid values of 0.011% ± 0.001% and 0.00158 ± 0.0004 mEq acid/g sample and purities of 99.5% ± 0.2% and 100.2% ± 0.4%. Thin-layer chromatography indicated a major spot and a slight trace impurity by one system and only a major spot by the second system. Gas chromatography using one system indicated one major peak and four impurities with a combined area of 3.0% relative to the major peak. Gas chromatography using another system indicated one major peak and four impurities with a combined area of 2.8% relative to the major peak. Another analysis of lot C090882 by gas chromatography using the second system indicated one major peak and three impurities with a combined area of 2.27% relative to the major peak. The overall purity of lot C090882 was determined to be greater than 97%.

For lot L-121 1-87, elemental analyses for carbon and hydrogen were in agreement with the theoretical values for butyl benzyl phthalate. Karl Fischer water analysis indicated 0.11% ± 0.01% water. Functional group titrations indicated a free acid value of 0.00112 ± 0.00003 mEq acid/g sample and a purity of 99.1% ± 0.4%. Thin-layer chromatography indicated a major spot and two slight trace impurities by one system and a major spot and one trace impurity by the second system. Gas chromatography using one system indicated one major peak and five impurities with a combined area of 1.20% relative to the major peak. Gas chromatography using another system indicated one major peak and three impurities with a combined area of 1.04% relative to the major peak. The overall purity of lot L-121 1-87 was determined to be greater than or equal to 99%.

Stability studies of the bulk chemical were performed by the analytical chemistry laboratory using gas chromatography. These studies indicated that butyl benzyl phthalate 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 4° C in the dark in tightly sealed 1-gallon amber-glass bottles during the 10-week modified mating study and the 26-week study and at approximately 22° C in 1-gallon amber-glass bottles with Teflon®-lined lids during the 2-year study.

Stability was monitored during the 10-week modified mating study, the 26-week study, and the 2-year study using gas chromatography. No degradation of the bulk chemical was detected.

## PREPARATION AND ANALYSIS OF DOSE FORMULATIONS

The dose formulations were prepared weekly by mixing butyl benzyl phthalate with feed (Table G1). Homogeneity and stability studies of 300, 3,000, 6,250, 24,000, and 25,000 ppm dose formulations were performed by the analytical chemistry laboratory and/or the study laboratory using high-performance liquid chromatography. Homogeneity was confirmed, and the stability of the dose formulations was confirmed for at least 4 weeks at 5° C when stored protected from light.

Periodic analyses of the dose formulations of butyl benzyl phthalate were conducted at the study laboratories and analytical chemistry laboratory using high-performance liquid chromatography. For the 10-week modified mating study, the 26-week study, and the 2-year study, the formulations were analyzed every 8 to 9 weeks (Tables G2 and G3). All of the dose formulations analyzed were within 10% of the target concentration. Results of periodic referee analyses performed by the analytical chemistry laboratory agreed with the results obtained by the study laboratory (Table G4).

## 10-WEEK MODIFIED MATING STUDY

Male and female F344/N rats were obtained from Simonsen Laboratories, Inc. (Gilroy, CA). On receipt, the male rats were 4 weeks old. Males were quarantined for 11 days and were 6 weeks old on the first day of the study. Females were 20 to 24 weeks old on the first day of the mating period. 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 sentinel rats using the protocols of the NTP Sentinel Animal Program (Appendix J).

Groups of 15 male rats were fed diets containing 0, 300, 2,800, or 25,000 ppm butyl benzyl phthalate for 10 weeks and then were allowed to recover for 2 days. Male rats were housed individually during the exposure and recovery periods, and two untreated females were housed with each male during the 7-day mating period. On the first day of vaginal plug or sperm detection (gestation day 0), females were housed individually. Feed and water were available *ad libitum*. Clinical findings were recorded daily for rats. Feed consumption by males was recorded weekly. The males were weighed initially, weekly, and at the end of the study. The females were weighed at the beginning of the 7-day mating period and on gestation days 0, 6, 9, and 13. Details of the study design and animal maintenance are summarized in Table 1.

At the end of the 10-week modified mating study, the males were necropsied and evaluated for reproductive toxicity. The parameters evaluated are listed in Table 1. Methods used were those described in the NTP's sperm morphology and vaginal cytology evaluations protocol (NTP, 1983b). The right testis and right epididymis were isolated, weighed, and fixed and preserved for histopathology. The tail of the epididymis (cauda epididymis) was then removed from the epididymal body (corpus epididymis) and weighed. Test yolk buffer was applied to slides, and a small incision was made at the distal border of the cauda epididymis. The sperm effluxing from the incision were dispersed in the buffer on the slides, and the numbers of motile and nonmotile spermatozoa were counted for five fields per slide by two observers. Following completion of the sperm motility estimates, each right cauda epididymis was placed in buffered saline solution. Caudae were finely minced, and the tissue was incubated in the saline solution, after which samples were removed for morphology evaluation, and the remainder were heat fixed at 65° C. Sperm density was then determined microscopically with the aid of a hemacytometer. Four sperm morphology slides were prepared for each animal evaluated. An aliquot of killed sperm suspension was stained in a test tube, spread on a microscope slide under a coverslip, and examined.

At the end of the 10-week modified mating study, all males were anesthetized with carbon dioxide, and blood was collected from the retroorbital sinus for hematology analyses. Blood for hematology determinations was placed in tubes containing potassium EDTA as the anticoagulant. Leukocyte, erythrocyte, and reticulocyte counts were determined from blood smears by light microscopy. The hematology parameters measured are listed in Table 1.

A necropsy was performed on all males after the 7-day mating period. The brain, heart, right kidney, liver, lung, prostate gland, seminal vesicle, right testis, and thymus were weighed. A necropsy was performed on all mated females on gestation day 13, and on all virgin females 13 days after the end of the mating period. The uterus and ovaries were stained with 10% ammonium sulfide and examined grossly. Table 1 lists the parameters examined. Histopathologic examinations were performed on 0 and 25,000 ppm male rats. In addition, the epididymis, prostate gland, seminal vesicle, and testis were examined in all other exposure groups. Table 1 lists the tissues and organs routinely examined.

## 26-Week Study

The 26-week study was conducted to evaluate the cumulative toxic effects of repeated exposure to butyl benzyl phthalate and to determine the appropriate doses to be used in the 2-year study.

Male F344/N rats were obtained from Simonsen Laboratories, Inc. (Gilroy, CA). On receipt, the rats were 4 weeks old. Animals were quarantined for 11 days and were 6 weeks old on the first day of the study. Before initiation of the study, five male rats were randomly selected for parasite evaluation and gross observation for evidence of disease. At 13 weeks and at the end of the study, serologic analyses were performed on five male sentinel rats using the protocols of the NTP Sentinel Animal Program (Appendix J).

Groups of 15 male rats were fed diets containing 0, 300, 900, 2,800, 8,300, or 25,000 ppm butyl benzyl phthalate. Feed and water were available *ad libitum*. Rats were housed five per cage. Clinical findings were recorded daily. Feed consumption was recorded weekly. The animals were weighed initially, weekly, and at the end of the study. Details of the study design and animal maintenance are summarized in Table 1.

At the end of the 26-week study, samples were collected from 0, 300, 8,300, and 25,000 ppm male rats for sperm morphology evaluations. The parameters evaluated are listed in Table 1. Methods used were the same as those described for males in the 10-week modified mating study.

On days 30, 60, 90, 120, 150, and at the end of the study, blood was collected from all rats (except for the 25,000 ppm males on days 120 and 150) from the retroorbital sinus for hematology analyses. The rats were anesthetized with carbon dioxide. Hematology parameters were determined using the same methods as described for males in the 10-week modified mating study. The hematology parameters measured are listed in Table 1.

A necropsy was performed on all rats. The brain, heart, right kidney, liver, lung, prostate gland, seminal vesicle, right testis, and thymus were weighed. Tissues for microscopic examination were fixed and preserved in 10% neutral buffered formalin, processed and trimmed, embedded in paraffin, sectioned to a thickness of 5 to 6 $\mu$m, and stained with hematoxylin and eosin. A complete histopathologic examination was performed on all 0 and 25,000 ppm rats. In addition, the epididymis, prostate gland, seminal vesicle, and testis were examined in all other exposure groups. Table 1 lists the tissues and organs routinely examined.

## 2-Year Study

### Study Design

Groups of 60 male rats were fed diets containing 0, 3,000, 6,000, or 12,000 ppm butyl benzyl phthalate, and groups of 60 female rats were fed diets containing 0, 6,000, 12,000, or 24,000 ppm butyl benzyl phthalate. Ten male and ten female rats from each group were bled at 6 months for hematology and at 6 and 8 months for hormone assays. After 15 months of exposure, 10 male and 10 female rats from each group were evaluated for hematology, hormone assays, organ weights, and histopathology.

### Source and Specification of Animals

Male and female F344/N rats were obtained from Simonsen Laboratories, Inc. (Gilroy, CA) for use in the 2-year study. Rats were 4 weeks old upon receipt and were quarantined for 11 days before the beginning of the study. Five male and five female rats were selected for parasite evaluation and gross observation for evidence of disease. Rats were approximately 6 weeks old at the beginning of the study. The health of the animals was monitored during the study according to the protocols of the

NTP Sentinel Animal Program (Appendix J). After approximately 1 year, an additional health check was performed on sentinel animals. All animals were positive for either pinworms or pinworm eggs.

## Animal Maintenance

Rats were housed five per cage. Feed and water were available *ad libitum*. Feed consumption was measured weekly for 12 weeks and monthly thereafter for controls and approximately every month for exposed groups. 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 the beginning of the study, weekly for 13 weeks, monthly thereafter, and at the end of the study.

Blood for hematology analyses was collected from all rats from the retroorbital sinus at 6 and 15 months. Blood for hormone assays was collected from all rats at 6 and 15 months and at study termination and from all males at 8 months. The rats were anesthetized with carbon dioxide or a mixture of carbon dioxide and oxygen. Blood for hematology determinations and plasma hormone assays was placed in tubes containing potassium EDTA as the anticoagulant. Blood for serum hormone assays was placed in tubes without anticoagulant, allowed to clot at room temperature, and centrifuged. Hematology parameters were measured on a Technicon H-1. Erythrocyte, reticulocyte, and differential leukocyte counts and morphologic evaluations of blood cells were determined from blood smears by light microscopy. Analyses for thyroxine, triiodothyronine, and testosterone were performed with radioimmunoassay kits (Diagnostic Products Corp., Los Angeles, CA). Analyses for thyroid-stimulating hormone and prolactin were performed with radioimmunoassay methods and reagents from the National Institute of Arthritis, Diabetes, Digestive, and Kidney Disease. Adrenocorticotropic hormone levels were measured using a double-antibody technique. The hematology and hormone assay parameters measured are listed in Table 1.

A complete necropsy and microscopic examination were performed on all rats. At the 15-month interim evaluation, the right epididymis, right kidney, liver, and right testis of rats 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 5 to 6 $\mu$m, 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. Tissues examined microscopically are listed in Table 1.

Microscopic evaluations were completed by the study laboratory pathologist, and the pathology data were entered into the Toxicology Data Management System. The 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 adrenal gland, kidney, liver, mammary gland (females), pancreas, pituitary gland (pars distalis), prostate gland (males), skin (males), spleen, thyroid gland (males), urinary bladder, and uterus (females) for all neoplastic and nonneoplastic lesions.

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 pathologists, 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., intestine, harderian gland, mammary gland, 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 was used, a procedure based on the overall proportion of affected animals.

### 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, hormone assay, spermatid, spermatozoal, fertility, and developmental toxicity 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 statistical 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). Fertility and developmental toxicity data were also analyzed using the Chi-square 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 26-week 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, the 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 butyl benzyl phthalate 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 mouse bone marrow cells, mutations in L5178Y mouse lymphoma cells, and sex-linked recessive lethal mutations in *Drosophila melanogaster*. The protocols for these studies and the results are given in Appendix C.

The genetic toxicity studies of butyl benzyl phthalate 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 Butyl Benzyl Phthalate**

| 10-Week Modified Mating Study | 26-Week Study | 2-Year Study |
|---|---|---|
| **Study Laboratory**<br>Hazleton Laboratories America, Inc. (Rockville, MD) | Hazleton Laboratories America, Inc. (Rockville, MD) | Southern Research Institute (Birmingham, AL) |
| **Strain and Species**<br>Rats: F344/N | Rats: F344/N | Rats: F344/N |
| **Animal Source**<br>Simonsen Laboratories, Inc. (Gilroy, CA) | Simonsen Laboratories, Inc. (Gilroy, CA) | Simonsen Laboratories, Inc. (Gilroy, CA) |
| **Time Held Before Studies**<br>11 days (males) | 11 days | 11 days |
| **Average Age When Studies Began**<br>6 weeks (males)<br>20 to 24 weeks (females at the beginning of the 7-day mating period) | 6 weeks | 6 weeks |
| **Date of First Dose**<br>24 September 1984 (males) | 24 September 1984 | 26 June 1989 |
| **Duration of Dosing**<br>10 weeks (males) | 26 weeks | 104 weeks (males),<br>105 weeks (females) |
| **Date of Last Dose**<br>2 December 1984 (males) | 25-26 March 1985 | 17 June (males) and<br>24 June (females) 1991 |
| **Necropsy Dates**<br>12-13 December (males) and<br>16-22 December (females) 1984 | 25-26 March 1985 | 15-Month interim evaluation -<br>26 September (males) and<br>27 September (females) 1990<br>Terminal -<br>24-26 June (males) and<br>1-3 July (females) 1991 |
| **Average Age at Necropsy**<br>16 weeks (males) and<br>22 to 26 weeks (females) | 32 weeks | 15-Month interim evaluation -<br>72 weeks<br>Terminal -<br>110 weeks (males) and<br>111 weeks (females) |
| **Size of Study Groups**<br>15 males and 30 untreated females | 15 males | 15-Month interim evaluation -<br>10 males and 10 females<br>Terminal -<br>50 males and 50 females |

TABLE 1
Experimental Design and Materials and Methods in the Feed Studies of Butyl Benzyl Phthalate
(continued)

| 10-Week Modified Mating Study | 26-Week Study | 2-Year Study |
|---|---|---|
| **Method of Distribution**<br>Animals were distributed randomly into groups of approximately equal mean body weights | Same as 10-week modified mating study | Same as 10-week modified mating study |
| **Animals per Cage**<br>1 (except during 7-day mating period, when one male was house with two females) | 5 | 5 |
| **Method of Animal Identification**<br>Toe clip and cage label | Toe clip and cage label | Tail tattoo |
| **Diet**<br>NIH-07 open formula meal diet (Zeigler Brothers, Inc., Gardners, PA), available *ad libitum* | Same as 10-week modified mating study | Same as 10-week modified mating study |
| **Water Distribution**<br>Tap water (Rockville municipal supply) via glass water bottles (Lab Products, Garfield, NJ; Ancare Corp., Manhasset, NY), available *ad libitum* | Same as 10-week modified mating study | Tap water (Birmingham municipal supply) via automatic watering system (Edstrom Industries, Inc., Waterford, WI), available *ad libitum* |
| **Cages**<br>Polycarbonate cages (Lab Products, Inc., Garfield, NJ), changed twice weekly | Same as 10-week modified mating study | Solid-bottom, polycarbonate cages (Lab Products, Inc., Maywood, NJ), changed twice weekly |
| **Bedding**<br>Sani-Chips® heat-treated hardwood chips (P.J. Murphy Forest Products Corp., Rochelle Park, NJ), changed twice weekly | Same as 10-week modified mating study | Sani-Chips® heat-treated hardwood chips (P.J. Murphy Forest Products Corp., Montville, NJ), changed twice weekly |
| **Cage/Rack Filters**<br>Nonwoven polyester cage filters (Snow Filtration Co., Cincinnati, OH), changed every 2 weeks | Same as 10-week modified mating study | Reemay® spun-bonded polyester rack filters (Andico, Birmingham, AL), changed every 2 weeks |
| **Racks**<br>Aluminum racks (Lab Products, Garfield, NJ), rotated every 2 weeks | Same as 10-week modified mating study | Stainless steel racks (Lab Products, Inc., Maywood, NJ), rotated every 2 weeks |

TABLE 1
**Experimental Design and Materials and Methods in the Feed Studies of Butyl Benzyl Phthalate**
(continued)

| 10-Week Modified Mating Study | 26-Week Study | 2-Year Study |
|---|---|---|
| **Animal Room Environment** | | |
| Temperature: 21° to 27° C<br>Relative humidity: 26% to 69%<br>Fluorescent light: 12 hours/day<br>Room air: 12 to 15 changes/hour | Temperature: 21° to 26° C<br>Relative humidity: 24% to 69%<br>Fluorescent light: 12 hours/day<br>Room air: 12 to 15 changes/hour | Temperature: 13.1° to 29.2° C<br>Relative humidity: 22.2% to 89.0%<br>Fluorescent light: 12 hours/day<br>Room air: minimum of 10 changes/hour |
| **Doses** | | |
| 0, 300, 2,800, or 25,000 ppm in feed, available *ad libitum* (males) | 0, 300, 900, 2,800, 8,300, or 25,000 ppm in feed, available *ad libitum* | 0, 3,000, 6,000, or 12,000 ppm (males) or 0, 6,000, 12,000, or 24,000 ppm (females) in feed, available *ad libitum* |
| **Type and Frequency of Observation** | | |
| Males were observed twice daily; males were weighed initially, weekly, and at the end of the study; clinical observations were recorded daily. Feed consumption by males was recorded weekly. | Observed twice daily; animals were weighed initially, weekly, and at the end of the study; clinical observations were recorded daily. Feed consumption was recorded weekly. | Observed twice daily; animals were weighed and clinical observations were recorded initially, weekly for 13 weeks, monthly thereafter, and at the end of the study. Feed consumption was recorded weekly for 12 weeks and monthly thereafter for controls and approximately monthly for exposed groups. |
| **Method of Sacrifice** | | |
| Carbon dioxide asphyxiation and exsanguination | Carbon dioxide asphyxiation and exsanguination | Carbon dioxide asphyxiation |
| **Necropsy** | | |
| Necropsy performed on all males. Organs weighed were brain, heart, right kidney, liver, lung, prostate gland, seminal vesicle, right testis, and thymus. | Necropsy performed on all rats. Organs weighed were brain, heart, right kidney, liver, lung, prostate gland, seminal vesicle, right testis, and thymus. | Necropsy performed on all rats. Organs weighed at the 15-month interim evaluation were right epididymis, right kidney, liver, and right testis. |

TABLE 1
Experimental Design and Materials and Methods in the Feed Studies of Butyl Benzyl Phthalate
(continued)

| 10-Week Modified Mating Study | 26-Week Study | 2-Year Study |
|---|---|---|
| **Clinical Pathology** | | |
| Blood for hematology was collected from the retroorbital sinus of all males surviving to the end of the study. ***Hematology:*** hematocrit, hemoglobin, erythrocyte and reticulocyte counts, mean cell volume, mean cell hemoglobin, mean cell hemoglobin concentration, platelet count, and total leukocyte count and differential. | Blood for hematology was collected from the retroorbital sinus on days 30, 60, 90, 120, 150, and at end of the study from all rats (except for 25,000 ppm males on days 120 and 150). ***Hematology:*** hematocrit, hemoglobin, erythrocyte and reticulocyte counts, mean cell volume, mean cell hemoglobin, mean cell hemoglobin concentration, platelet counts, and total leukocyte counts and differentials. | Blood was collected from all rats from the retroorbital sinus at 6 and 15 months for hematology, and from males at 6, 8, and 15 months and at study termination, and from females at 6 and 15 months and at study termination for hormone assays. ***Hematology:*** hematocrit; hemoglobin; erythrocyte, reticulocyte, and nucleated erythrocyte counts; mean cell volume; mean cell hemoglobin; mean cell hemoglobin concentration; platelet counts; total leukocyte counts and differentials. ***Hormone assays:*** thyroid-stimulating hormone, triiodothyronine (6 and 15 months and study termination only in males), thyroxine (6 and 15 months and study termination only in males), adrenocorticotropic hormone (6 and 15 months and study termination only in males), testosterone, and prolactin (15 months and study termination only in females) |
| **Histopathology** | | |
| Complete histopathology was performed on all 0 and 25,000 ppm males. In addition to gross lesions and tissue masses, the tissues examined included: adrenal gland, brain, esophagus, heart, large intestine (cecum, colon, rectum), small intestine (duodenum, jejunum, ileum), kidney, liver, lung (and mainstem bronchi), lymph nodes (mandibular and mesenteric), mammary gland, nose, pancreas, parathyroid gland, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular), testis (with epididymis and seminal vesicle), thymus, thyroid gland, trachea, and urinary bladder. The testis (with epididymis and seminal vesicle) and prostate gland were also examined in all other groups of rats. | Complete histopathology was performed on all 0 and 25,000 ppm rats. In addition to gross lesions and tissue masses, the tissues examined included: adrenal gland, brain, esophagus, heart, large intestine (cecum, colon, rectum), small intestine (duodenum, jejunum, ileum), kidney, liver, lung (and mainstem bronchi), lymph nodes (mandibular and mesenteric), mammary gland, nose, pancreas, parathyroid gland, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular), testis (with epididymis and seminal vesicle), thymus, thyroid gland, trachea, and urinary bladder. The testis (with epididymis and seminal vesicle) and prostate gland were also examined in all other groups of rats. | Complete histopathology was performed on all groups of rats. In addition to gross lesions and tissue masses, the tissues examined included: adrenal gland, brain (three sections), clitoral gland, femur (and bone marrow), heart, large intestine (cecum, colon, rectum), small intestine (duodenum, jejunum, ileum), kidney, liver, lung (and mainstem bronchi), lymph nodes (mandibular and mesenteric), mammary gland, oral mucosa (and tongue), ovary, pancreas, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular), testes (with epididymis and seminal vesicle), thymus, thyroid gland, urinary bladder, and uterus. |

TABLE 1
**Experimental Design and Materials and Methods in the Feed Studies of Butyl Benzyl Phthalate**
(continued)

| 10-Week Modified Mating Study | 26-Week Study | 2-Year Study |
|---|---|---|
| **Reproductive Tissue Evaluations** | | |
| At the end of the study, sperm samples were collected from all male rats for sperm count, motility, and morphology evaluations. The right cauda, epididymis, and testis were weighed. | At the end of the study, sperm samples were collected from male rats in the 0, 300, 8,300, and 25,000 ppm groups for sperm count, motility, and morphology evaluations. The right cauda, epididymis, and testis were weighed. | None |
| **Fertility, Maternal Body Weight, and Developmental Toxicity Evaluations** | | |
| Females were observed twice daily during mating and gestation. Females were weighed at the beginning of the 7-day mating period and on gestation days 0, 6, 9, and 13. Necropsies were performed on mated females on gestation day 13, and on virgin females 13 days after the end of the mating period. The uterus and ovaries were examined. The parameters evaluated included: male and female fertility indexes, maternal body weights, live and dead fetuses, resorptions, corpora lutea, and gross abnormalities. | None | None |

# RESULTS

## RATS

### 10-WEEK MODIFIED MATING STUDY

All rats survived to the end of the study (Table 2). The final mean body weight and body weight gain of the 25,000 ppm males were significantly less than those of the controls.

Feed consumption by the 300 and 2,800 ppm groups was similar to that by the controls. Feed consumption by the 25,000 ppm group was less than that by the controls at the end of the study. Dietary levels of 300, 2,800, and 25,000 ppm delivered average daily doses of approximately 20, 200, and 2,200 mg butyl benzyl phthalate/kg body weight. No clinical findings were considered to be related to butyl benzyl phthalate exposure.

TABLE 2
Survival, Body Weights, and Feed Consumption of Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate

| Dose (ppm) | Survival[a] | Mean Body Weight[b] (g) | | | Final Weight Relative to Controls (%) | Feed Consumption[c] | |
|---|---|---|---|---|---|---|---|
| | | Initial | Final | Change | | Week 2 | Week 12 |
| 0 | 15/15 | 110 ± 3 | 320 ± 4 | 211 ± 2 | | 14 | 16 |
| 300 | 15/15 | 111 ± 3 | 319 ± 3 | 208 ± 4 | 100 | 14 | 16 |
| 2,800 | 15/15 | 111 ± 3 | 316 ± 3 | 205 ± 3 | 99 | 14 | 16 |
| 25,000 | 15/15 | 110 ± 3 | 226 ± 4** | 116 ± 3** | 71 | 22 | 11 |

** Significantly different (P≤0.01) from the control group by Williams' or Dunnett's test.
[a] Number of animals surviving at 10 weeks/number initially in group
[b] Weights and weight changes are given as mean ± standard error.
[c] Feed consumption is expressed as grams of feed consumed per animal per day.

A few minimal hematology changes occurred in the 25,000 ppm male rats (Table E1). There was some evidence of a minimal anemia; this was characterized by a decreased erythrocyte count. The substantially lower final mean body weight of the 25,000 ppm males suggests, however, that these rats were not eating or drinking as much as the controls, and dehydration could have occurred. Dehydration can cause a relative erythrocytosis due to decreased blood volume and hemoconcentration. If significant dehydration occurred in these rats, the severity of the anemia could have been masked. The erythrocytes were macrocytic, evidenced by an increase in mean cell volume. Increases in mean cell volume usually suggest increased numbers of larger, immature erythrocytes (reticulocytes) in the circulation. However, the reticulocyte count was not increased in 25,000 ppm males, indicating the decrease in erythrocyte count was too minimal to stimulate a strong bone marrow response. An increase in mean cell hemoglobin occurred and would be consistent with the larger erythrocyte size. The platelet count was increased and would be consistent with a reactive thrombocytosis. Reactive thrombocytosis can be caused by a variety of conditions, including bone marrow response to anemia.

The absolute and relative testis and prostate gland weights of the 25,000 ppm males were significantly less than those of the controls (Table D1). The other lower organ weights of rats in the 25,000 ppm group were attributed to the moderately lower mean body weight gain of this group. There were marked degenerative testicular and associated epididymal degenerative changes in the 25,000 ppm males. All changes were associated with damage primarily to the germinal epithelium, which resulted in necrosis (detritus) and loss (atrophy and hypospermia) of germinal epithelium and with formation of syncytia of spermatids (giant cells) (Table 3).

**TABLE 3**
**Incidences of Selected Nonneoplastic Lesions in Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate**

| Dose | 0 ppm | 300 ppm | 2,800 ppm | 25,000 ppm |
|---|---|---|---|---|
| Testis[a] | 15 | 15 | 15 | 15 |
| Seminiferous Tubule, Atrophy[b] | 0 | 1 (4.0)[c] | 0 | 15** (4.0) |
| Seminiferous Tubule, Giant Cells | 0 | 0 | 0 | 10** (1.3) |
| Seminiferous Tubule, Necrosis | 0 | 0 | 0 | 3 (1.0) |
| Epididymis | 15 | 15 | 15 | 15 |
| Hypospermia | 0 | 1 (4.0) | 0 | 15** (4.0) |
| Tail, Chronic Inflammation | 0 | 0 | 0 | 4* (1.3) |
| Tail, Detritus | 0 | 0 | 0 | 11** (2.9) |

* Significantly different ($P \leq 0.05$) from the control group by the Fisher exact test
** $P \leq 0.01$
[a] Number of animals with organ examined microscopically
[b] Number of animals with lesion
[c] Average severity grade of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked

The absolute right cauda, right epididymis, and right testis weights of the 25,000 ppm males were significantly less than those of the controls (Table F1). The epididymal spermatozoal concentrations in 2,800 and 25,000 ppm males were significantly less than that in the controls. Sperm motility and epididymal spermatozoal parameters were not calculated for males exposed to 25,000 ppm due to the absence of sperm, and those in other groups of exposed males were similar to those in the controls. Although 10 females mated to 25,000 ppm males were initially found to be sperm positive, none of these females were pregnant at necropsy. The fertility indices of males and females in the 25,000 ppm group were significantly lower than those of the controls (Table F2). The maternal body weights of females mated to 300 and 2,800 ppm males were similar to those of females mated to control males. There were no maternal clinical findings related to exposure of males to butyl benzyl phthalate. There were no significant differences in litter data between the controls and the 300 and 2,800 ppm groups.

## 26-Week Study

Two control males, one 300 ppm male, one 900 ppm male, one 2,800 ppm male, and four 25,000 ppm males died during the study (Table 4). All deaths occurred during anesthetization for the collection of blood from the retroorbital sinus. The final mean body weight and body weight gain of the 25,000 ppm males were significantly less than those other controls. Except for the 25,000 ppm males, feed consumption by all exposed groups was similar to that by the controls. Feed consumption was not measured for the 25,000 ppm group because there was excessive scattering of the feed at this exposure concentration. Dietary levels of 300, 900, 2,800, and 8,300 ppm delivered average daily doses of approximately 30, 60, 180, and 550 mg butyl benzyl phthalate/kg body weight. No clinical findings were related to butyl benzyl phthalate exposure.

TABLE 4
Survival, Body Weights, and Feed Consumption of Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate

| Dose (ppm) | Survival[a] | Mean Body Weight[b] (g) Initial | Final | Change | Final Weight Relative to Controls (%) | Feed Consumption[c] Week 2 | Week 28 |
|---|---|---|---|---|---|---|---|
| 0 | 13/15[d] | 113 ± 3 | 365 ± 15 | 251 ± 14 | | 18 | 16 |
| 300 | 14/15[e] | 113 ± 3 | 391 ± 5 | 278 ± 4 | 107 | 25 | 17 |
| 900 | 14/15[f] | 112 ± 2 | 403 ± 6 | 290 ± 5 | 110 | 17 | 16 |
| 2,800 | 14/15[g] | 112 ± 3 | 374 ± 8 | 260 ± 6 | 102 | 16 | 15 |
| 8,300 | 15/15 | 113 ± 3 | 375 ± 8 | 263 ± 6 | 103 | 17 | 15 |
| 25,000 | 11/15[h] | 112 ± 3 | 254 ± 13** | 140 ± 10** | 70 | —[i] | — |

** Significantly different (P≤0.01) from the control group by Williams' or Dunnett's test
a Number of animals surviving at 26 weeks/number initially in group
b Weights and weight changes are given as mean ± standard error. Subsequent calculations are based on animals surviving to the end of the study.
c Feed consumption is expressed as grams of feed consumed per animal per day.
d Week of death: 5, 9
e Week of death: 5
f Week of death: 18
g Week of death: 9
h Week of death: 5, 9, 9, 13
i No feed consumption data were calculated for this exposure group.

The hematology data are listed in Table E2. An exposure-related macrocytic responsive anemia was present in the 25,000 ppm group at all time points evaluated. The anemia was characterized by a minimal to mild decrease in hematocrit values and/or erythrocyte counts. Additionally, minimal erythrocyte count decreases occurred sporadically in the 2,800 and 8,300 ppm groups at various time points. The macrocytosis, evidenced by a minimal to mild increase in mean cell volume, would suggest increased numbers of circulating reticulocytes. Reticulocyte counts were increased on days 60 and 90 and would be consistent with a hematopoietic response to the anemia. An increase in mean cell hemoglobin occurred in the 8,300 and 25,000 ppm rats at all time points. In general, increases in mean cell hemoglobin would be a reflection of the larger erythrocyte size. There were minimal increases in the mean cell hemoglobin concentration in the 8,300 and 25,000 ppm exposure groups. Increases in mean cell hemoglobin concentration have been related to erythrocyte hemolysis (*in vivo* or *in vitro*) or

alterations in hemoglobin concentrations or hematocrit values. Differences in other hematology variables did not exhibit an exposure-related response and were not considered relevant.

The absolute and relative testis weights of the 25,000 ppm males were significantly less than those of the controls (Table D2). Other organ weight differences in rats were attributed to changes in body weight gains or were not considered to be related to exposure to butyl benzyl phthalate. The absolute right cauda and right epididymis weights and the sperm concentration of 25,000 ppm males were significantly less than those of the controls (Table F3). Sperm motility and epididymal spermatozoal parameters were not calculated for males exposed to 25,000 ppm, and those in other exposed groups of males were similar to those in the controls. The incidences of hypospermia and of atrophy of the seminiferous tubule in the testis and of hypospermia in the epididymis in 25,000 ppm males were significantly greater than those of the controls (Table 5). The degenerative changes of the testis and epididymis of the 25,000 ppm males were qualitatively and quantitatively similar to those observed in males in the 10-week modified mating study.

**TABLE 5**
**Incidences of Selected Nonneoplastic Lesions in Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate**

| Dose | 0 ppm | 300 ppm | 900 ppm | 2,800 ppm | 8,300 ppm | 25,000 ppm |
|---|---|---|---|---|---|---|
| Testis[a] | 15 | 15 | 15 | 15 | 15 | 15 |
| Seminiferous Tubule, Atrophy[b] | 0 | 0 | 0 | 1 (1.0)[c] | 0 | 15** (3.9) |
| Seminiferous Tubule, Giant Cells | 0 | 0 | 0 | 0 | 0 | 5* (2.2) |
| Seminiferous Tubule, Hypospermia | 0 | 0 | 0 | 0 | 0 | 15** (4.0) |
| Epididymis | 15 | 15 | 15 | 15 | 15 | 15 |
| Hypospermia | 0 | 0 | 0 | 0 | 0 | 15** (4.0) |
| Tail, Detritus | 0 | 0 | 0 | 1 (1.0) | 0 | 13** (2.2) |

* Significantly different ($P \leq 0.05$) from the control group by the Fisher exact test
** $P \leq 0.01$
[a] Number of animals with organ examined microscopically
[b] Number of animals with lesion
[c] Average severity of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked

*Dose Selection Rationale:* Based on lower mean body weights and increased incidences of nonneoplastic lesions in the 25,000 ppm rats, butyl benzyl phthalate exposure concentrations selected for male rats in the 2-year study were 0, 3,000, 6,000, or 12,000 ppm. In a previous 2-year NTP study, the only evidence of chronic toxicity or carcinogenicity in female rats was a marginal increase in the incidence of mononuclear cell leukemia. A higher exposure concentration of 24,000 ppm was added for females in this 2-year study to maximize the potential for observing adverse chemical effects. Therefore, exposure concentrations of 0, 6,000, 12,000, or 24,000 ppm were selected for females in the 2-year study.

## 2-YEAR STUDY

### *Survival*

Estimates of 2-year survival probabilities for male and female rats are shown in Table 6 and in the Kaplan-Meier survival curves (Figure 1). Survival of all exposed groups of males and females was similar to that of the controls.

### *Body Weights, Feed and Compound Consumption, and Clinical Findings*

The mean body weight of the 12,000 ppm males was 4% to 10% less than that of the controls throughout most of the study, and the mean body weight of the 24,000 ppm females was 7% to 27% less than that of the controls throughout most of the study. Mean body weights of all 3,000 and 6,000 ppm males and of 6,000 and 12,000 ppm females were similar to those of the controls throughout the study (Tables 7 and 8, Figure 2). Feed consumption by the females exposed to 24,000 ppm was less than that by the controls at the beginning of the study, but was similar to that by the controls by week 6 (Tables H1 and H2). Feed consumption by all other exposed groups of males and females was similar to that by the controls. Dietary levels of 3,000, 6,000, or 12,000 ppm delivered average daily doses of approximately 120, 240, or 500 mg butyl benzyl phthalate/kg body weight to males. Dietary levels of 6,000, 12,000, or 24,000 ppm delivered average daily doses of approximately 300, 600, or 1,200 mg/kg to females. There were no clinical findings in exposed groups of rats related to butyl benzyl phthalate exposure.

### *Hematology and Hormone Assays*

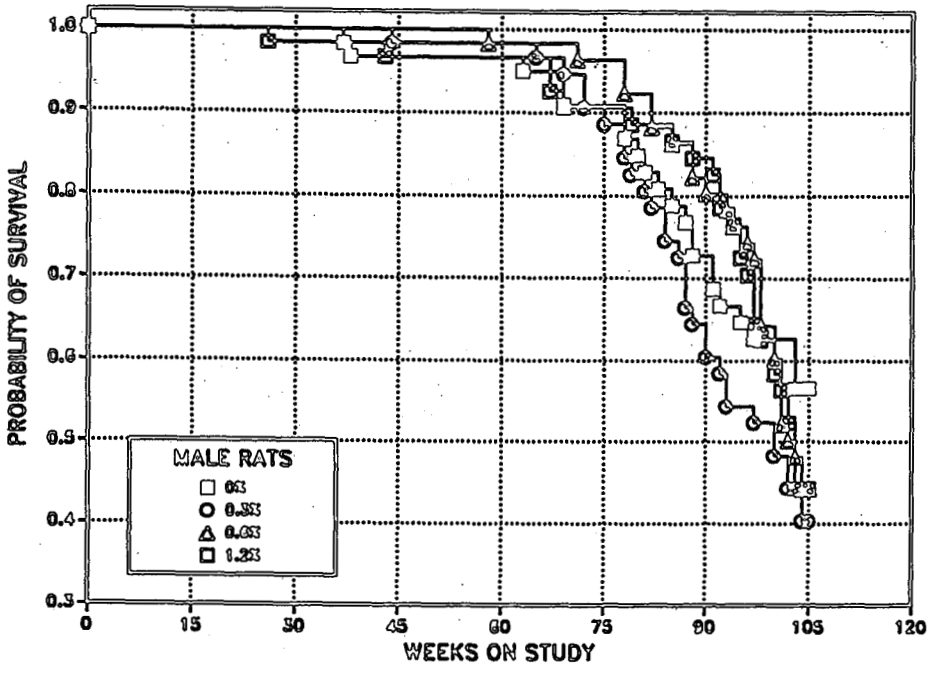
The hematology and hormone assay data for rats evaluated at 6, 8, and 15 months and at the end of the study are listed in Table E3. In general, hematology changes were sporadic and minor. At 6 months, a minimal decrease in erythrocyte count and an increase in mean cell hemoglobin, similar to that which occurred in the 26-week study, occurred in male rats in the 12,000 ppm group. In female rats, a decreased hematocrit value occurred at 15 months in the 24,000 ppm group. At this time point, however, the mean cell volume was decreased, indicating that smaller than normal red cells (microcytosis) were being released from the bone marrow. A microcytic anemia is usually associated with a reduced erythropoiesis and may be caused by a variety of conditions including decreased nutritional status. The marked mean body weight decrease of the 24,000 ppm female rats would be consistent with decreased nutrition. There was also a mild decrease in triiodothyronine concentrations in the 24,000 ppm females at 6 and 15 months and at study termination. No alterations in the thyroxine and thyroid-stimulating hormone concentrations occurred, suggesting that the decrease in triiodothyronine was related to a nonthyroidal disorder. In nonthyroidal disorders, decreases in circulating triiodothyronine concentrations have been attributed to the decreased conversion of thyroxine to triiodothyronine peripherally (e.g. liver) and can be related to numerous conditions including poor nutrition. It appears that thyroid hormones decrease in extrathyroidal tissues (except for the brain) as a response to situations in which preservation of energy is advantageous, and the triiodothyronine decrease observed in various nonthyroidal disorders is probably related to caloric deprivation or conservation.

**TABLE 6**
**Survival of Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Male** | | | | |
| Animals initially in study | 60 | 60 | 60 | 60 |
| 15-Month interim evaluation[a] | 10 | 10 | 10 | 10 |
| Moribund | 19 | 19 | 26 | 26 |
| Natural deaths | 3 | 11 | 2 | 2 |
| Animals surviving to study termination | 28 | 20 | 22 | 22 |
| Percent probability of survival at end of study[b] | 57 | 40 | 44 | 44 |
| Mean survival (days)[c] | 625 | 617 | 644 | 632 |
| Survival analysis[d] | P=0.794 | P=0.185 | P=0.570 | P=0.520 |

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Female** | | | | |
| Animals initially in study | 60 | 60 | 60 | 60 |
| 15-Month interim evaluation[a] | 10 | 10 | 10 | 10 |
| Moribund | 23 | 17 | 16 | 20 |
| Natural deaths | 2 | 4 | 5 | 1 |
| Animals surviving to study termination | 25 | 29 | 29 | 29 |
| Percent probability of survival at end of study | 50 | 59 | 58 | 59 |
| Mean survival (days) | 651 | 651 | 651 | 644 |
| Survival analysis | P=0.770N | P=0.567N | P=0.596N | P=0.759N |

[a] Censored from survival analyses
[b] Kaplan-Meier determinations
[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 exposed columns. A negative trend or lower mortality in an exposure group is indicated by N.



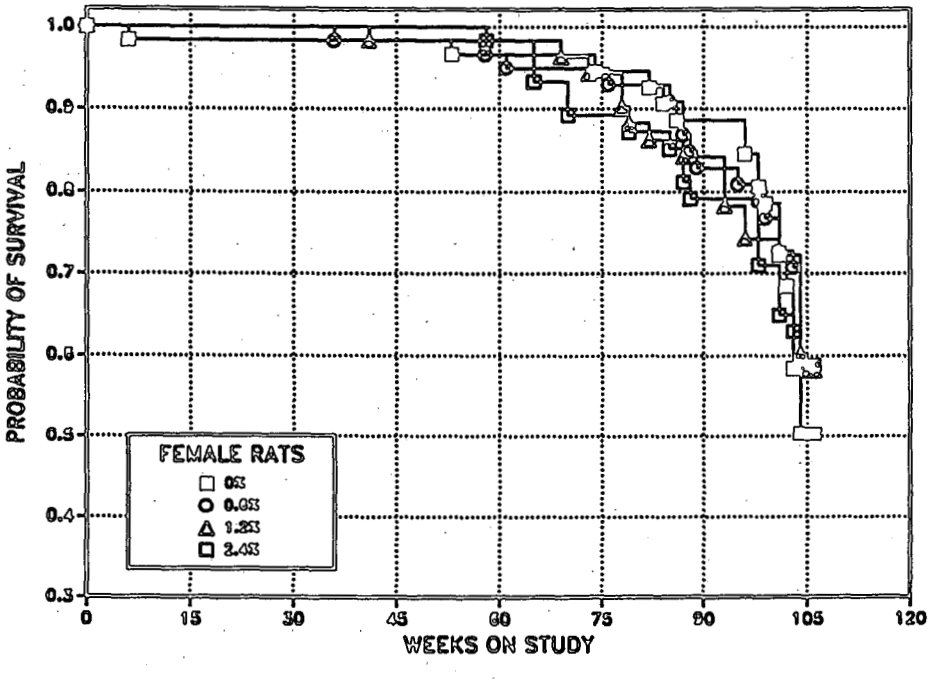




FIGURE 1
Kaplan-Meier Survival Curves for Rats Administered Butyl Benzyl Phthalate in Feed for 2 Years

TABLE 7
**Mean Body Weights and Survival of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| Weeks on Study | 0 ppm Av. Wt. (g) | 0 ppm No. of Survivors | 3,000 ppm Av. Wt. (g) | 3,000 ppm Wt. (% of controls) | 3,000 ppm No. of Survivors | 6,000 ppm Av. Wt. (g) | 6,000 ppm Wt. (% of controls) | 6,000 ppm No. of Survivors | 12,000 ppm Av. Wt. (g) | 12,000 ppm Wt. (% of controls) | 12,000 ppm No. of Survivors |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 113 | 60 | 111 | 98 | 60 | 109 | 97 | 60 | 108 | 96 | 60 |
| 2 | 149 | 60 | 148 | 99 | 60 | 147 | 99 | 60 | 141 | 95 | 60 |
| 3 | 183 | 60 | 186 | 102 | 60 | 184 | 101 | 60 | 180 | 98 | 60 |
| 4 | 205 | 60 | 208 | 102 | 60 | 209 | 102 | 60 | 208 | 101 | 60 |
| 5 | 233 | 60 | 236 | 101 | 60 | 233 | 100 | 60 | 226 | 97 | 60 |
| 6 | 253 | 60 | 255 | 101 | 60 | 253 | 100 | 60 | 243 | 96 | 60 |
| 7 | 266 | 60 | 267 | 100 | 60 | 258 | 97 | 60 | 255 | 96 | 60 |
| 8 | 286 | 60 | 287 | 100 | 60 | 274 | 96 | 60 | 272 | 95 | 60 |
| 9 | 298 | 60 | 299 | 100 | 60 | 292 | 98 | 60 | 284 | 95 | 60 |
| 10 | 310 | 60 | 310 | 100 | 60 | 301 | 97 | 60 | 291 | 94 | 60 |
| 11 | 319 | 60 | 320 | 100 | 60 | 307 | 96 | 60 | 301 | 94 | 60 |
| 12 | 326 | 60 | 328 | 101 | 60 | 319 | 98 | 60 | 312 | 96 | 60 |
| 13 | 339 | 60 | 338 | 100 | 60 | 321 | 95 | 60 | 315 | 93 | 60 |
| 17 | 370 | 60 | 367 | 99 | 60 | 350 | 95 | 60 | 338 | 91 | 60 |
| 21 | 391 | 60 | 389 | 100 | 60 | 367 | 94 | 60 | 351 | 90 | 60 |
| 25 | 391 | 60 | 403 | 103 | 60 | 381 | 98 | 60 | 362 | 93 | 60 |
| 29 | 416 | 60 | 411 | 99 | 60 | 394 | 95 | 60 | 377 | 91 | 59 |
| 33 | 423 | 60 | 421 | 100 | 60 | 399 | 95 | 60 | 382 | 90 | 59 |
| 37 | 429 | 60 | 429 | 100 | 60 | 410 | 96 | 60 | 389 | 91 | 59 |
| 41 | 437 | 58 | 434 | 99 | 60 | 422 | 96 | 60 | 394 | 90 | 59 |
| 45 | 443 | 58 | 451 | 102 | 59 | 429 | 97 | 60 | 405 | 92 | 58 |
| 49 | 449 | 58 | 449 | 100 | 59 | 433 | 97 | 60 | 413 | 92 | 58 |
| 53 | 451 | 58 | 451 | 100 | 59 | 437 | 97 | 60 | 414 | 92 | 58 |
| 57 | 450 | 58 | 453 | 101 | 59 | 432 | 96 | 60 | 412 | 92 | 58 |
| 61 | 451 | 58 | 445 | 99 | 59 | 434 | 96 | 59 | 413 | 92 | 58 |
| 65 | 451 | 57 | 446 | 99 | 59 | 434 | 96 | 59 | 420 | 93 | 58 |
| 69[a] | 446 | 46 | 445 | 100 | 47 | 440 | 99 | 49 | 419 | 94 | 46 |
| 73 | 448 | 45 | 447 | 100 | 45 | 441 | 99 | 48 | 427 | 95 | 45 |
| 77 | 446 | 45 | 443 | 99 | 44 | 441 | 99 | 48 | 428 | 96 | 45 |
| 81 | 440 | 42 | 437 | 99 | 41 | 431 | 98 | 46 | 422 | 96 | 44 |
| 85 | 440 | 40 | 431 | 98 | 37 | 424 | 96 | 44 | 418 | 95 | 44 |
| 89 | 439 | 36 | 430 | 98 | 32 | 428 | 98 | 41 | 413 | 94 | 42 |
| 93 | 440 | 33 | 429 | 97 | 28 | 422 | 96 | 40 | 422 | 96 | 39 |
| 97 | 433 | 32 | 428 | 99 | 27 | 410 | 95 | 37 | 411 | 95 | 35 |
| 101 | 426 | 31 | 419 | 98 | 24 | 407 | 96 | 29 | 402 | 94 | 29 |
| **Mean for weeks** | | | | | | | | | | | |
| 1-13 | 252 | | 253 | 100 | | 247 | 98 | | 241 | 96 | |
| 14-52 | 417 | | 417 | 100 | | 398 | 95 | | 379 | 91 | |
| 53-101 | 443 | | 439 | 99 | | 429 | 97 | | 417 | 94 | |

[a] Interim evaluation occurred during week 66.

TABLE 8
Mean Body Weights and Survival of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate

| Weeks on Study | 0 ppm | | 6,000 ppm | | | 12,000 ppm | | | 24,000 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 | 99 | 60 | 98 | 99 | 60 | 96 | 97 | 60 | 92 | 93 | 60 |
| 2 | 121 | 60 | 121 | 101 | 60 | 122 | 101 | 60 | 114 | 94 | 60 |
| 3 | 135 | 60 | 135 | 100 | 60 | 134 | 99 | 60 | 127 | 94 | 60 |
| 4 | 142 | 60 | 145 | 102 | 60 | 142 | 100 | 60 | 137 | 97 | 60 |
| 5 | 153 | 60 | 151 | 99 | 60 | 149 | 97 | 60 | 142 | 93 | 60 |
| 6 | 159 | 59 | 162 | 102 | 60 | 157 | 99 | 60 | 151 | 95 | 60 |
| 7 | 167 | 59 | 167 | 100 | 60 | 163 | 98 | 60 | 157 | 94 | 60 |
| 8 | 171 | 59 | 171 | 100 | 60 | 167 | 97 | 60 | 160 | 94 | 60 |
| 9 | 175 | 59 | 176 | 100 | 60 | 171 | 98 | 60 | 166 | 95 | 60 |
| 10 | 178 | 59 | 179 | 101 | 60 | 175 | 98 | 60 | 171 | 96 | 60 |
| 11 | 180 | 59 | 183 | 102 | 60 | 176 | 98 | 60 | 170 | 95 | 60 |
| 12 | 186 | 59 | 187 | 100 | 60 | 181 | 97 | 60 | 175 | 94 | 60 |
| 13 | 185 | 59 | 187 | 101 | 60 | 179 | 97 | 60 | 176 | 95 | 60 |
| 17 | 198 | 59 | 197 | 100 | 60 | 193 | 98 | 60 | 183 | 93 | 60 |
| 21 | 204 | 59 | 204 | 100 | 60 | 196 | 97 | 60 | 187 | 92 | 60 |
| 25 | 213 | 59 | 213 | 100 | 60 | 206 | 97 | 60 | 193 | 91 | 60 |
| 29 | 218 | 59 | 219 | 100 | 60 | 211 | 97 | 60 | 197 | 91 | 60 |
| 33 | 223 | 59 | 225 | 101 | 60 | 216 | 97 | 60 | 199 | 89 | 60 |
| 37 | 231 | 59 | 231 | 100 | 59 | 224 | 97 | 60 | 203 | 88 | 60 |
| 41 | 238 | 59 | 239 | 100 | 59 | 230 | 97 | 60 | 206 | 87 | 60 |
| 45 | 240 | 59 | 244 | 102 | 59 | 239 | 99 | 59 | 207 | 86 | 60 |
| 49 | 258 | 59 | 261 | 101 | 59 | 248 | 96 | 59 | 212 | 82 | 60 |
| 53 | 269 | 59 | 270 | 100 | 59 | 258 | 96 | 59 | 214 | 80 | 60 |
| 57 | 276 | 58 | 275 | 100 | 59 | 266 | 96 | 59 | 214 | 78 | 60 |
| 61 | 281 | 58 | 280 | 99 | 58 | 270 | 96 | 59 | 216 | 77 | 59 |
| 65 | 286 | 58 | 287 | 100 | 57 | 277 | 97 | 59 | 220 | 77 | 59 |
| 69[a] | 296 | 48 | 293 | 99 | 47 | 281 | 95 | 48 | 222 | 75 | 46 |
| 73 | 298 | 48 | 301 | 101 | 47 | 284 | 95 | 48 | 226 | 76 | 44 |
| 77 | 300 | 47 | 304 | 101 | 46 | 287 | 96 | 47 | 230 | 77 | 44 |
| 81 | 300 | 47 | 303 | 101 | 46 | 291 | 97 | 44 | 222 | 74 | 43 |
| 85 | 312 | 45 | 308 | 99 | 46 | 299 | 96 | 43 | 231 | 74 | 43 |
| 89 | 321 | 44 | 321 | 100 | 41 | 306 | 95 | 42 | 237 | 74 | 39 |
| 93 | 320 | 44 | 321 | 100 | 41 | 308 | 96 | 39 | 236 | 74 | 39 |
| 97 | 319 | 42 | 323 | 101 | 40 | 309 | 97 | 37 | 233 | 73 | 39 |
| 101 | 318 | 36 | 325 | 102 | 36 | 307 | 97 | 36 | 232 | 73 | 32 |
| **Mean for weeks** | | | | | | | | | | | |
| 1-13 | 158 | | 159 | 101 | | 155 | 98 | | 149 | 94 | |
| 14-52 | 225 | | 226 | 100 | | 218 | 97 | | 199 | 88 | |
| 53-101 | 300 | | 301 | 100 | | 288 | 96 | | 226 | 75 | |

[a] Interim evaluation occurred during week 66.

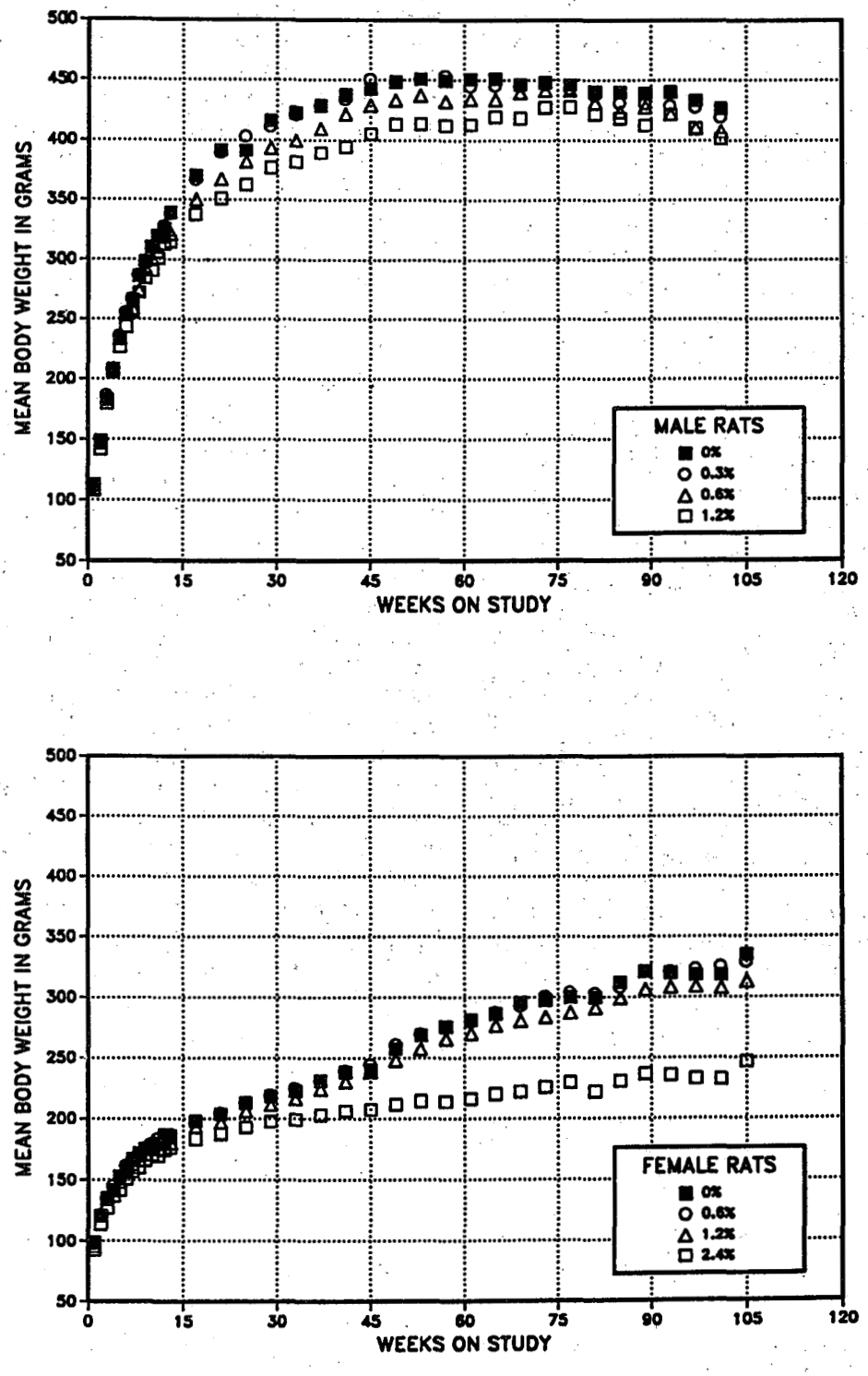



FIGURE 2
Growth Curves for Rats Administered Butyl Benzyl Phthalate in Feed for 2 Years

### *Pathology and Statistical Analyses*

This section describes the statistically significant or biologically noteworthy changes in the incidences of neoplasms and/or nonneoplastic lesions of the pancreas, urinary bladder, kidney, skin, preputial gland, mammary gland, pituitary gland, and other organs. 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.

*Pancreas:* At 2 years, the incidences of pancreatic acinar cell adenoma and pancreatic acinar cell adenoma or carcinoma (combined) in 12,000 ppm males were significantly greater than those in the controls (Tables 9 and A3). The incidences of adenoma and of adenoma or carcinoma (combined) in this group exceeded the ranges of historical controls from NTP 2-year feed studies (Tables 9 and A4a). One carcinoma was observed in a 12,000 ppm male (Tables 9 and A1). This neoplasm has never been observed in the historical controls from NTP 2-year feed studies (Tables 9 and A4a). At 2 years, the incidence of focal hyperplasia of the pancreatic acinar cell in 12,000 ppm males was also significantly greater than that in the controls (Tables 9 and A5).

The foci of acinar cell hyperplasia were variably circumscribed and often minimally compressed the surrounding parenchyma. The glandular pattern within the hyperplasias was pronounced, and the cells were slightly enlarged compared to normal with mitotic figures occasionally observed. Generally, the adenomas were more discrete and were larger (greater than 3 mm in diameter) with greater compression of the surrounding parenchyma. Also, the acinar cell pattern was more heterogeneous with greater mitotic activity. The carcinoma was larger and more pleomorphic than the adenomas.

Two pancreatic acinar cell adenomas were observed in the 24,000 ppm females (Tables 9 and B1). While the incidence of this neoplasm (4%) was within the range of historical controls from NTP 2-year feed studies (Tables 9 and B4a), pancreatic neoplasms have only been observed in 3 of 1,194 females in the historical database. Hyperplasia, adenoma, and carcinoma of the pancreatic acinar cell represent a morphological and biological continuum with the morphological distinction between hyperplasias and adenomas not always clearly apparent. There were no increases in the incidences of hyperplasia or carcinoma in exposed females. However, because pancreatic neoplasms are rare in control groups, these lesions were classified as adenomas, and because a pancreatic tumorigenic effect occurred in the males, the pancreatic adenomas in the 24,000 ppm females were considered to be possibly related to exposure to butyl benzyl phthalate.

TABLE 9
**Incidences of Neoplasms and Nonneoplastic Lesions of the Pancreas in Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| Dose | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Male** | | | | |
| Number Examined Microscopically | 50 | 49 | 50 | 50 |
| Acinus, Focal Hyperplasia[a] | 4 (2.5)[b] | 7 (2.1) | 9 (2.3) | 12 (2.3) |
| Acinus, Adenoma[c] | | | | |
| Overall rate[d] | 3/50 (6%) | 2/49 (4%) | 3/50 (6%) | 10/50 (20%) |
| Adjusted rate[e] | 10.7% | 7.4% | 13.6% | 41.0% |
| Terminal rate[f] | 3/28 (11%) | 1/20 (5%) | 3/22 (14%) | 8/22 (36%) |
| First incidence (days) | 729 (T) | 569 | 729 (T) | 709 |
| Logistic regression test[g] | P=0.006 | P=0.543N | P=0.548 | P=0.016 |
| Acinus, Carcinoma[h] | | | | |
| Overall rate | 0/50 (0%) | 0/49 (0%) | 0/50 (0%) | 1/50 (2%) |
| Acinus, Adenoma or Carcinoma[i] | | | | |
| Overall rate | 3/50 (6%) | 2/49 (4%) | 3/50 (6%) | 11/50 (22%) |
| Adjusted rate | 10.7% | 7.4% | 13.6% | 42.7% |
| Terminal rate | 3/28 (11%) | 1/20 (5%) | 3/22 (14%) | 8/22 (36%) |
| First incidence (days) | 729 (T) | 569 | 729 (T) | 674 |
| Logistic regression test | P=0.003 | P=0.543N | P=0.548 | P=0.014 |

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Female** | | | | |
| Number Examined Microscopically | 50 | 50 | 50 | 50 |
| Acinus, Focal Hyperplasia | 1 (3.0) | 4 (2.5) | 2 (2.5) | 0 |
| Acinus, Adenoma[j] | | | | |
| Overall rate | 0/50 (0%) | 0/50 (0%) | 0/50 (0%) | 2/50 (4%) |

(T)Terminal sacrifice
* Significantly different (P≤0.05) from the control by the logistic regression test
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: 19/1,191 (1.6% ± 2.4%); range, 0%-10%
d Number of animals with neoplasm per number of animals with pancreas 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. A lower incidence in an exposure group is indicated by N.
h Historical incidence: 0/1,919 (0.0%)
i Historical incidence: 19/1,191 (1.6% ± 2.4%); range, 0%-10%
j Historical incidence: 2/1,194 (0.2% ± 0.8%); range, 0%-4%

*Urinary Bladder:* At 2 years, transitional epithelial papillomas in the urinary bladder were observed in one control female and in two 24,000 ppm females (Tables 10 and B1). The incidence of this neoplasm exceeded the range of historical controls from NTP 2-year feed studies (Tables 10 and B4b). At 2 years, the incidence of transitional epithelial hyperplasia in 24,000 ppm females was significantly greater than in the controls (Tables 10 and B5).

The hyperplasias consisted of mild to moderate, focal to multifocal thickening of the transitional epithelium of the urinary bladder. The papillomas projected into the lumen of the urinary bladder and were characterized by papillary fronds that were covered with proliferative transitional epithelium and supported by a fibrous stalk. Hyperplasia and papillomas of the urinary bladder represent a morphological and biological continuum in the progression of proliferative lesions, and, although the two papillomas were small, they appeared to represent the next step in the progression of the observed hyperplasias. The occurrence of two papillomas and the supportive increase in the incidence of hyperplasia in the 24,000 ppm females suggest that these lesions may have been associated with exposure to butyl benzyl phthalate.

TABLE 10
Incidences of Neoplasms and Nonneoplastic Lesions of the Urinary Bladder in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate

| Dose | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| Number Examined Microscopically | 50 | 50 | 50 | 50 |
| Hyperplasia, Transitional Epithelium[a] | 4 (1.5)[b] | 0 | 1 (4.0) | 10* (2.2) |
| Papilloma, Transitional Epithelium[c] | 1 | 0 | 0 | 2 |

* Significantly different ($P \leq 0.05$) from the control by the logistic regression test
[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: 4/1,182 (0.3% ± 0.8%); range, 0%-2%

*Kidney:* At the 15-month interim evaluation, the absolute right kidney weight of the 12,000 ppm females and the relative kidney weights of all exposed groups of males and the 24,000 ppm females were significantly greater than those of the controls (Table D3). The severities of renal tubule pigmentation in 12,000 ppm males and in 24,000 ppm females were greater than those in the controls at 15 months (Table 11). A marginal increase in severity was observed in 12,000 ppm males at 2 years. The pigment was yellowish-brown, coarsely granular, and present predominantly within the cytoplasm of tubule epithelial cells. Special stains performed on kidneys from four animals were positive for hemosiderin (Prussian Blue) and marginally positive for ceroid (AFB), suggesting an exacerbation of the pigment normally observed in aging rats. At 2 years, the incidences of mineralization of the kidney in 6,000 and 24,000 ppm females were significantly less than that in the controls (Tables 11 and B5), and the severity was decreased in exposed females at 15 months and 2 years. The incidences of nephropathy in 6,000, 12,000, and 24,000 ppm females and of transitional epithelial hyperplasia of the kidney in 12,000 ppm females were significantly greater than those in the controls (Tables 11 and B5).

Minimal to mild mineralization of the pars recta of the kidney is a common finding in female F344/N rats; the mineralization is often intraluminal, but may be intracellular. There were also slight increases in the incidences of nephropathy in exposed groups of

females. Nephropathy is extremely common in aging F344/N rats (particularly males) and was apparently marginally exacerbated in this study. Though these effects are possibly related to exposure to butyl benzyl phthalate, they are marginal changes of common background lesions and are of minimal biological significance. Also, there was an increase in the incidence of hyperplasia of the renal pelvic transitional epithelium in 12,000 ppm females. These hyperplasias were characterized by focal minimal thickening of the epithelium, which often contained a central focus of mineralization.

TABLE 11
**Incidences of Nonneoplastic Lesions of the Kidney in Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| Dose | 0 ppm | | 3,000 ppm | | 6,000 ppm | | 12,000 ppm | |
|---|---|---|---|---|---|---|---|---|
| **Male** | | | | | | | | |
| **15-Month Interim Evaluation** | | | | | | | | |
| Number Examined Microscopically | 10 | | 10 | | 10 | | 10 | |
| Nephropathy[a] | 10 | (1.4)[b] | 10 | (1.3) | 10 | (1.3) | 9 | (1.1) |
| Renal Tubule, Pigmentation | 10 | (1.0) | 10 | (1.0) | 10 | (1.2) | 10 | (2.0) |
| **2-Year Study** | | | | | | | | |
| Number Examined Microscopically | 50 | | 50 | | 50 | | 50 | |
| Mineralization | 0 | | 1 | (1.0) | 2 | (1.5) | 0 | |
| Nephropathy | 48 | (2.1) | 47 | (2.2) | 50 | (2.3) | 48 | (2.0) |
| Renal Tubule, Pigmentation | 49 | (1.5) | 48 | (1.5) | 50 | (1.6) | 50 | (1.9) |
| Transitional Epithelium, Hyperplasia | 6 | (1.0) | 10 | (1.3) | 6 | (1.2) | 1 | (2.0) |
| | **0 ppm** | | **6,000 ppm** | | **12,000 ppm** | | **24,000 ppm** | |
| **Female** | | | | | | | | |
| **15-Month Interim Evaluation** | | | | | | | | |
| Number Examined Microscopically | 10 | | 10 | | 10 | | 10 | |
| Mineralization | 10 | (2.5) | 9 | (1.8) | 9 | (1.6) | 8 | (1.6) |
| Nephropathy | 7 | (1.0) | 10 | (1.0) | 10 | (1.0) | 10 | (1.0) |
| Renal Tubule, Pigmentation | 10 | (1.4) | 10 | (1.9) | 10 | (1.8) | 10 | (2.0) |
| **2-Year Study** | | | | | | | | |
| Number Examined Microscopically | 50 | | 50 | | 50 | | 50 | |
| Mineralization | 43 | (2.1) | 34* | (1.7) | 37 | (1.4) | 35* | (1.4) |
| Nephropathy | 34 | (1.4) | 47** | (1.1) | 43* | (1.1) | 45** | (1.0) |
| Renal Tubule, Pigmentation | 49 | (1.4) | 49 | (1.5) | 49 | (1.4) | 47 | (1.6) |
| Transitional Epithelium, Hyperplasia | 0 | | 3 | (1.0) | 7** | (1.0) | 4 | (1.0) |

* Significantly different ($P \leq 0.05$) from the control by the logistic regression test
** $P \leq 0.01$
[a] Number of animals with lesion
[b] Average severity of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked

*Skin:* At 2 years, the incidences of acanthosis and hyperkeratosis of the skin in 12,000 ppm males were significantly greater than in the controls (Tables 12 and A5). The identification of these lesions was based upon gross observations of crusty areas involving the skin of the tail. Microscopically, there was mild to moderate thickening (acanthosis) of the epidermis with excessive overlying keratin (keratinization). The pathogenesis of this minimal effect is uncertain.

TABLE 12
Incidences of Nonneoplastic Lesions of the Skin in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate

| Dose | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| Number Necropsied | 50 | 50 | 50 | 50 |
| Acanthosis[a] | 0 | 2 (3.0)[b] | 2 (2.5) | 10** (2.6) |
| Hyperkeratosis | 0 | 2 (3.0) | 3 (3.0) | 13** (3.1) |

** Significantly different ($P \leq 0.01$) from the control by the logistic regression test
[a] Number of animals with lesion
[b] Average severity of lesions in affected animals: 1 = minimal; 2 = mild; 3 = moderate; 4 = marked

*Preputial Gland:* At 2 years, the incidences of preputial gland adenomas in males occurred with a negative trend (Tables 13 and A3). The incidence of adenoma or carcinoma (combined) of the preputial gland in 12,000 ppm males was significantly less than in the controls, and the incidences occurred with a negative trend (Tables 13 and A3). The incidence of preputial gland adenoma or carcinoma (combined) in 12,000 ppm males was less than the range of historical controls in NTP 2-year feed studies (Tables 13 and A4b). In no recent control group has the incidence been 0% as in the 12,000 ppm males in this study. The incidences of hyperplasia of the preputial gland in exposed groups were similar to that in the controls (Tables 13 and A5). A decrease in the incidence of clitoral gland neoplasms, which is the comparable tissue in females, was not observed (Table B1). Xenobiotics that affect one of these tissues often affect the other, but not always. It is not clear whether this marginal decrease in the incidence of preputial gland neoplasms was related to exposure to butyl benzyl phthalate.

TABLE 13
**Incidences of Neoplasms and Nonneoplastic Lesions of the Preputial Gland in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| Dose | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| Number Examined Microscopically | 50 | 50 | 50 | 50 |
| Hyperplasia[a] | 1 (3.0)[b] | 2 (2.0) | 2 (2.5) | 1 (2.0) |
| Adenoma[c] | | | | |
| Overall rate[d] | 4/50 (8%) | 3/50 (6%) | 1/50 (2%) | 0/50 (0%) |
| Adjusted rate[e] | 12.8% | 15.0% | 4.5% | 0.0% |
| Terminal rate[f] | 2/28 (7%) | 3/20 (15%) | 1/22 (5%) | 0/22 (0%) |
| First incidence (days) | 644 | 729 (T) | 729 (T) | —[h] |
| Logistic regression test[g] | P=0.022N | P=0.593N | P=0.165N | P=0.059N |
| Adenoma or Carcinoma[i] | | | | |
| Overall rate | 5/50 (10%) | 8/50 (16%) | 2/50 (4%) | 0/50 (0%) |
| Adjusted rate | 15.1% | 33.9% | 9.1% | 0.0% |
| Terminal rate | 2/28 (7%) | 5/20 (25%) | 2/22 (9%) | 0/22 (0%) |
| First incidence (days) | 614 | 626 | 729 (T) | — |
| Logistic regression test | P=0.006N | P=0.201 | P=0.196N | P=0.032N |

(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: 84/1,164 (7.2% ± 5.6%); range, 0%-24%
[d] Number of animals with neoplasm per number of animals with preputial gland 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. A negative trend or lower incidence in an exposure group is indicated by N.
[h] Not applicable; no neoplasms in animal group
[i] Historical incidence: 124/1,164 (10.7% ± 7.5%); range, 2%-30%

*Mammary Gland:* At 2 years, the incidence of fibroadenoma of the mammary gland was significantly decreased in the 24,000 ppm females (Tables 14 and B3). The incidences of mammary gland fibroadenoma in the 6,000 and 12,000 ppm females only slightly exceeded the range of historical controls from NTP 2-year feed studies (Tables 14 and B4c). Fibroadenomas of the mammary gland are benign neoplasms that occur spontaneously at a high rate in female F344/N rats.

TABLE 14
Incidences of Neoplasms of the Mammary Gland in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate

| Dose | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **15-Month Interim Evaluation** | | | | |
| Number Necropsied | 10 | 10 | 10 | 10 |
| Fibroadenoma[a] | 0 | 1 | 1 | 0 |
| **2-Year Study** | | | | |
| Number Necropsied | 50 | 50 | 50 | 50 |
| Fibroadenoma[b] | | | | |
| Overall rate[c] | 28/50 (56%) | 30/50 (60%) | 31/50 (62%) | 11/50 (22%) |
| Adjusted rate[d] | 71.0% | 72.8% | 71.7% | 28.9% |
| Terminal rate[e] | 14/25 (56%) | 18/29 (62%) | 17/29 (59%) | 5/29 (17%) |
| First incidence (days) | 587 | 606 | 509 | 487 |
| Logistic regression test[f] | P<0.001N | P=0.379 | P=0.318 | P<0.001N |

[a] Number of animals with neoplasm
[b] Historical incidence for 2-year NTP feed studies with untreated controls: 465/1,202 (38.7% ± 12.7%); range, 8%-58%
[c] Number of animals with neoplasm per number of animals necropsied
[d] Kaplan-Meier estimated neoplasm incidence at the end of the study after adjustment for intercurrent mortality
[e] Observed incidence in animals surviving until the end of the study
[f] 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. A negative trend or lower incidence in an exposure group is indicated by N.

*Pituitary Gland:* At 2 years, the incidences of adenoma or carcinoma (combined) of the pars distalis of the pituitary gland in females occurred with a negative trend (0 ppm, 22/49; 6,000 ppm, 26/50; 12,000 ppm, 25/48; 24,000 ppm, 13/50; Table B3). Incidences of pituitary gland neoplasms in F344/N rats have been demonstrated to correlate with body weight (Seilkop, 1995). It is probable that the decrease in the incidences of these neoplasms in the 24,000 ppm females is associated with the significantly lower mean body weights of this group.

*Other Organs:* In addition to the effects mentioned above, there were other statistically significant increased or decreased incidences of nonneoplastic lesions. In males, there were increases in the incidences of granuloma in the liver (0 ppm, 0/50; 3,000 ppm, 0/50; 6,000 ppm, 0/50; 12,000 ppm, 7/50; Table A5) and of corpora amylacae (mineralization) in the prostate gland (17/50, 28/50, 32/50, 30/50); and a decrease in the incidence of bile duct hyperplasia (39/50, 41/50, 38/50, 27/50). In females, there were decreases in the incidences of focal hypertrophy of the adrenal cortex (0 ppm, 9/50; 6,000 ppm, 11/50; 12,000 ppm, 7/50; 24,000 ppm, 0/50; Table B5) and of cytoplasmic vacuolization of hepatocytes (7/50, 6/50, 2/50, 0/50). In general, these nonneoplastic lesions may occur spontaneously in high numbers and represent relatively insignificant changes within the individual organs and/or tissues. While the potential contribution of exposure to butyl benzyl phthalate and/or lower mean body weights as causal factors of these effects remains undetermined, the biological importance of the changes is considered minimal.

## GENETIC TOXICOLOGY

Butyl benzyl phthalate was not mutagenic in a series of *in vitro* tests, but positive responses were obtained in two *in vivo* studies conducted with male mice.

Butyl benzyl phthalate was tested in two laboratories for mutagenicity in *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1537 with and without Aroclor-induced S9 liver enzymes (Zeiger *et al.*, 1985; Table C1). Negative results were obtained in both laboratories in all strains using concentrations of butyl benzyl phthalate that were the highest permitted by experimental design. *In vitro* studies with mammalian cell systems also gave negative results with and without S9. No induction of trifluorothymidine resistance was obtained in L5178Y mouse lymphoma cells with concentrations of butyl benzyl phthalate that formed stable solutions (Myhr and Caspary, 1991; Table C2). Increases in mutant colonies were observed in the absence of S9 in cultures treated with concentrations that produced precipitation, but such responses were not considered valid by experimental quality control parameters. Therefore, the test was concluded to be negative. No induction of sister chromatid exchanges or chromosomal aberrations was observed in cultured Chinese hamster ovary cells treated with butyl benzyl phthalate (Galloway *et al.*, 1987; Tables C3 and C4). In the first sister chromatid exchange trial without S9, the response was considered to be equivocal due to a significant trend ($P=0.004$) in the absence of a significant increase in sister chromatid exchanges at any one dose level. However, results of the second trial were clearly negative, as was the single trial conducted with S9, and the test results were concluded to be negative overall.

No induction of sex-linked recessive lethal mutations was observed in germ dells of male *Drosophila melanogaster* administered 500 ppm butyl benzyl phthalate by injection or up to 50,000 ppm in dosed feed (Valencia *et al.*, 1985; Table C5).

*In vivo* studies with butyl benzyl phthalate in male mice gave positive results for induction of sister chromatid exchanges (Table C6) and chromosomal aberrations (Table C7) at standard sampling times over a dose range of 1,250 to 5,000 mg/kg. In the sister chromatid exchange test, a single trial with a sample time of 23 hours post-injection yielded a positive trend when the highest dose was excluded from the analysis ($P=0.0067$) because of a reduction in response at 5,000 mg/kg. The sister chromatid exchange test conducted with a 42-hour sample time also gave a weakly positive response by trend analysis. Neither trial was repeated. Positive trend analyses were obtained in each of two trials in the chromosomal aberrations test that used the standard harvest time of 17 hours post injection. The only significant dose in each trial was the highest dose (5,000 mg/kg). The single chromosomal aberrations trial that used a delayed harvest time of 36 hours showed no increases in chromosomal aberrations at any of the three dose levels tested.

In conclusion, butyl benzyl phthalate showed no evidence of mutagenicity in any of four *in vitro* tests performed with and without S9 activation enzymes. In addition, a test for induction of sex-linked recessive lethal mutations in germ cells of male *Drosophila melanogaster* gave negative results. Positive results were obtained in mouse bone marrow tests for sister chromatid exchanges and induction of chromosomal aberrations. Responses were weak, however, and the sister chromatid exchange test was not repeated.

# DISCUSSION AND CONCLUSIONS

Butyl benzyl phthalate is widely used either directly as a plasticizer in a variety of commercial polymers or indirectly as a chemical intermediate. Consequently, there is human exposure in occupational settings and through a variety of consumer products including food, cosmetics, furniture and interior finishings, numerous household items, and product packaging. Butyl benzyl phthalate was originally nominated for chronic evaluation in rats and mice by the U.S. Environmental Protection Agency as part of a class study of phthalates. A previous study of butyl benzyl phthalate in rats and mice resulted in unexplained chemical-related mortality in male rats beginning at about week 14 of the study (NTP, 1982a). Therefore, the present studies were conducted only in rats.

Since male reproductive morphology and function have been affected by other phthalate esters, a 10-week modified mating study was conducted to assess the possibility that butyl benzyl phthalate could also adversely affect male reproduction. Following exposure to butyl benzyl phthalate in feed for 10 weeks, marked chemical-related effects on male reproductive parameters included decreased testis, epididymis, and seminiferous tubule weights accompanied by dramatically decreased sperm concentrations. It is likely that these effects were related to the marked testicular degeneration seen in the 10-week modified mating study and the 26-week feed study.

The 26-week study in males was designed to help characterize the chemical-related mortality observed in the previously conducted NTP studies (NTP, 1982a). In those studies, internal hemorrhaging was suspected at necropsy, but it was not confirmed microscopically. There were no chemical-related deaths in the present 26-week study and, in general, hematology changes were minimal. The most prominent change was a macrocytic responsive anemia in the 25,000 ppm male rats. A cause for the anemia was not readily evident, but chronic occult hemorrhaging could account for the responsive anemia. Evidence of an anemia was also present in the 10-week modified mating study, but it was not as prominent as that in the 26-week study.

In the 2-year study, there was evidence of an anemia in the 24,000 ppm female rats. However, it was a microcytic, rather than a macrocytic, anemia and was probably related to the markedly lower mean body weight gain (decreased nutritional status) of this group. The 24,000 ppm female rats also had lower triiodothyronine concentrations. This is probably also related to the lower mean body weight (hence, decreased nutrition).

The exposure concentrations used for males in the present 2-year study (3,000, 6,000, and 12,000 ppm) were based on results from the 10-week modified mating study and the 26-week study. In each of these studies, exposure concentrations up to 25,000 ppm did not cause any chemical-related deaths, but the 25,000 ppm concentration did result in 30% lower final mean body weights. However, the 8,300 ppm concentration had no adverse effects on mean body weights in the 26-week study, suggesting that the males could tolerate higher exposure concentrations. Consequently, 12,000 ppm was selected as the highest exposure concentration for males in the 2-year study. The exposure concentrations selected for females were based on the previous 2-year study conducted by the NTP (1982a) in which exposure concentrations of 0, 6,000, and 12,000 ppm did not affect survival of exposed groups of females, and mean body weights in exposed groups were slightly lower than those of the controls throughout the study. The only evidence of chronic toxicity or carcinogenicity in the previous 2-year study was a marginal increase in the incidence of mononuclear cell leukemia in the 12,000 ppm female rats. Consequently, these exposure concentrations were used for males in this study, and a higher exposure concentration of 24,000 ppm was added for females to maximize the potential for observing adverse chemical effects. Therefore, the exposure concentrations used for females in this study were 6,000, 12,000, and 24,000 ppm.

For male and female rats, the selected exposure concentrations had no effect on mortality or morbidity. Mean body weights of 3,000 and 6,000 ppm males and of 6,000 and 12,000 ppm females were similar to those of the controls. The exposure of males to 12,000 ppm butyl benzyl phthalate resulted in 4% to 10% lower mean body weights throughout most of the study, whereas the exposure of females to 24,000 ppm resulted in a more severe mean body weight depression of about 7% to 27% throughout the study. Although the survival of 24,000 ppm females was similar to that of the controls, the degree of mean body weight depression suggests that a lower exposure concentration could have been used. The selected exposure concentrations did allow adequate numbers of animals to be evaluated for chronic toxicity and carcinogenicity, and they also ranged from concentrations causing no effects to those which resulted in chemical-related adverse effects.

Chemical-related neoplasms and nonneoplastic lesions were observed in rats in the present 2-year study. Exposure to butyl benzyl phthalate resulted in marked increases in the incidences of pancreatic acinar cell hyperplasia and adenoma in 12,000 ppm male rats. In the F344/N rat, these lesions are generally characterized by a clear morphological continuum from focal acinar cell hyperplasia to adenoma to carcinoma. Ten adenomas and one carcinoma were observed in the 12,000 ppm males (11/50, 22%). This was statistically significant (P=0.014) by logistic regression analysis, which is appropriate for this typically nonfatal neoplasm. This incidence is well above the average incidence and well outside the range reported in the historical database from NTP 2-year feed studies. Although only one carcinoma was observed in a 12,000 ppm male, no carcinomas have ever been observed in NTP historical control rats from feed studies. With the exception of an increased incidence of pancreatic acinar cell hyperplasia in 12,000 ppm males, there were no other lesions at this site in male rats that could be attributed to the administration of butyl benzyl phthalate.

Even though the females were administered exposure concentrations that were twice as high as those administered to males, only two pancreatic acinar cell adenomas were observed in the 24,000 ppm group. These were probably related to exposure to butyl benzyl phthalate because of the rarity of this neoplasm. This difference in susceptibility between males and females has been reported for another peroxisome proliferator (hydrochlorofluorocarbon 123) (Malley *et al.*, 1995) and appears to be consistent with results of investigations showing that testosterone is stimulatory and estrogen is inhibitory for growth of acinar cell neoplasms in rat models (Lhoste *et al.*, 1987a,b; Sumi *et al.*, 1989; Longnecker and Sumi, 1990). Other peroxisome proliferators such as clofibrate and nafenopin have caused pancreatic acinar cell neoplasms, but these chemicals were only evaluated in male rats (Reddy and Qureshi, 1979; Svoboda and Azarnoff, 1979).

The biological significance of the marginal increase in the incidence of transitional epithelial papilloma in the urinary bladder of female rats is uncertain. While this neoplasm was observed in two 24,000 ppm females versus one in the control females, the appearance of this neoplasm in the exposed groups was associated with increases in the incidence and severity of transitional epithelial hyperplasia. This suggests that these lesions may have been associated with exposure to butyl benzyl phthalate.

Butyl benzyl phthalate did not cause liver neoplasms. Liver neoplasms would be expected because they have been observed with other peroxisome proliferators. In collaborative studies conducted concomitantly with the present 2-year study at the same laboratory, butyl benzyl phthalate was evaluated for its ability to induce hepatic peroxisomes in female F344/N rats as evidenced by the activities of two hepatic peroxisomal enzyme markers, palmitoyl CoA oxidase and carnitine acetyl transferase, when evaluated after 1 month and 1 year of exposure to butyl benzyl phthalate (Monsanto, 1994). It appears that butyl benzyl phthalate can cause a mild yet detectable increase in peroxisome proliferation, although at a lower concentration than that induced by only 3 weeks of exposure to the positive control, di(2-ethylhexyl)-phthalate (Table 15).

TABLE 15
Enzyme Activities in Female Rats Administered Butyl Benzyl Phthalate or Di(2-ethylhexyl)phthalate in Feed[a]

| | Butyl Benzyl Phthalate | | | | Di(2-ethylhexyl)phthalate[b] |
|---|---|---|---|---|---|
| Dose | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm | 12,000 ppm |
| **1 Month** | | | | | |
| n | 5 | 5 | 5 | 5 | 5 |
| Palmitoyl CoA Oxidase (nmol/min/mg) | 20.7 ± 5.6 | 22.2 ± 3.4 | 34.8 ± 10.6* | 74.5 ± 18.2* | 100.2 ± 38.4* |
| Carnitine Acetyl Transferase (nmol/min/mg) | 5.0 ± 1.2 | 21.1 ± 3.9* | 36.8 ± 13.6* | 59.9 ± 17.2* | 83.7 ± 28.5* |
| **1 Year** | | | | | |
| n | 10 | 10 | 10 | 10 | 5 |
| Palmitoyl CoA Oxidase (nmol/min/mg) | 28.79 ± 5.56 | 33.11 ± 3.81 | 48.35 ± 10.57* | 71.24 ± 11.94* | 86.30 ± 12.24* |
| Carnitine Acetyl Transferase (nmol/min/mg) | 17.13 ± 6.89 | 26.21 ± 4.18* | 45.29 ± 6.82* | 64.44 ± 14.06* | 72.20 ± 10.46* |

* Significantly different ($P \leq 0.05$) from the control group by the two-tailed Student's *t*-test
[a] Mean ± standard deviation. Study performed at Southern Research Institute. A detailed description of the protocol and these data are presented by Monsanto (1994).
[b] Di(2-ethylhexyl)phthalate was a positive control administered only for the last 3 weeks of the study.

These peroxisome proliferation results agree with work by other investigators. An association of hepatic peroxisome proliferation (induced by structurally dissimilar hypolipidemic drugs and certain phthalate ester plasticizers) with rodent liver tumorigenicity has been demonstrated by Reddy *et al.* (1980, 1986), who suggested that the hepatocarcinogenicity of these chemicals can be correlated with their ability to induce peroxisome proliferation and cyanide-insensitive palmitoyl CoA oxidase. In a ranking of seven phthalate esters and several common hypolipidemic agents by induction of normalized enzyme activity, the palmitoyl CoA oxidase value of butyl benzyl phthalate was two, compared to one for aspirin, 15 for di(2-ethylhexyl)phthalate, and 29 to 304 for hypolipidemic agents such as ciprofibrate and WY-14643 that are known peroxisome proliferators and liver carcinogens in rodents (Reddy and Lalwai, 1983; Barber *et al.*, 1987). Critical analysis of the literature on over 70 chemicals that induce peroxisome proliferation in rodent livers reveals a near perfect correlation between peroxisome proliferation and the eventual appearance of hepatocarcinogenicity (Ashby *et al.*, 1994). *In vivo* and *in vitro* investigations failed to provide evidence for significant peroxisome proliferation in the human liver by the chemicals studied. Furthermore, epidemiology studies have failed to demonstrate an increased incidence of tumors in humans exposed chronically to therapeutic hypolipidemic drugs known to cause peroxisome proliferation in rodents (reviewed by Ashby *et al.*, 1994).

Interestingly, there was no increase in the incidence of mononuclear cell leukemia in female rats as was observed previously, although the present study used exposure concentrations that were twice as high as those administered in the previous study (NTP, 1982a).

The decreased incidence of mammary gland neoplasms in female rats was observed previously (NTP, 1982a) and is attributable to lower mean body weights in the exposed groups. Using the NTP historical

control database, a logistic regression model for predicting mammary gland neoplasm incidence was developed based on survival and body weight at 1 year (Seilkop, 1995). Application of this model to the butyl benzyl phthalate data revealed that the predicted neoplasm incidence of 20.6% for the 24,000 ppm female exposure group agrees with the observed incidence (11/50, 22%). Thus, the chemical-related decrease in the incidence of this neoplasm appears to be due primarily to the lower mean body weights.

Butyl benzyl phthalate showed no evidence of mutagenicity in four standard *in vitro* tests (Zeiger *et al.*, 1985; Galloway *et al.*, 1987; Myhr and Caspary, 1991), and a germ cell mutation test in *Drosophila* gave negative results (Valencia *et al.*, 1985). Weak but statistically significant increases were, however, observed in mouse bone marrow tests for sister chromatid exchanges and chromosomal aberrations. The highest dose used in these tests was 5,000 mg/kg, which is approximately four times the highest dose used in these studies (1,200 mg/kg, 25,000 ppm). It was only at the highest dose (5,000 mg/kg) that increases in the chromosomal aberrations were observed *in vivo* in male mice.

## CONCLUSIONS

Under the conditions of this 2-year feed study, there was *some evidence of carcinogenic activity** of butyl benzyl phthalate in male F344/N rats based on the increased incidences of pancreatic acinar cell adenoma and of acinar cell adenoma or carcinoma (combined). There was *equivocal evidence of carcinogenic activity* of butyl benzyl phthalate in female F344/N rats based on the marginally increased incidences of pancreatic acinar cell adenoma and of transitional epithelial papilloma of the urinary bladder.

Exposure of rats to butyl benzyl phthalate in feed for 2 years resulted in focal hyperplasia in the pancreas in male rats and in transitional epithelial hyperplasia in the urinary bladder of female rats.

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* 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.

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# APPENDIX A
# SUMMARY OF LESIONS IN MALE RATS IN THE 2-YEAR FEED STUDY OF BUTYL BENZYL PHTHALATE


| | | |
|---|---|---|
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TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Disposition Summary** | | | | |
| Animals initially in study | 60 | 60 | 60 | 60 |
| ***15-Month interim evaluation*** | 10 | 10 | 10 | 10 |
| Early deaths | | | | |
| Moribund | 19 | 19 | 26 | 26 |
| Natural deaths | 3 | 11 | 2 | 2 |
| Survivors | | | | |
| Terminal sacrifice | 28 | 20 | 22 | 22 |
| Animals examined microscopically | 60 | 60 | 60 | 60 |
| ***15-Month Interim Evaluation*** | | | | |
| **Endocrine System** | | | | |
| Adrenal medulla | (10) | (10) | (10) | (10) |
| Pheochromocytoma benign | 1 (10%) | | 1 (10%) | |
| Pituitary gland | (9) | (10) | (10) | (10) |
| Pars distalis, adenoma | 2 (22%) | 2 (20%) | | 1 (10%) |
| Thyroid gland | (10) | (10) | (10) | (10) |
| C-cell, adenoma | | | | 1 (10%) |
| **Genital System** | | | | |
| Testes | (10) | (10) | (10) | (10) |
| Bilateral, interstitial cell, adenoma | 3 (30%) | 3 (30%) | 2 (20%) | 5 (50%) |
| Interstitial cell, adenoma | 4 (40%) | 2 (20%) | 7 (70%) | 4 (40%) |
| **Hematopoietic System** | | | | |
| Spleen | (10) | (10) | (10) | (10) |
| Histiocytic sarcoma | 1 (10%) | | | |
| **Integumentary System** | | | | |
| Skin | (10) | (10) | (10) | (10) |
| Keratoacanthoma | 1 (10%) | | | |
| **Systemic Lesions** | | | | |
| Multiple organs[b] | (10) | (10) | (10) | (10) |
| Histiocytic sarcoma | 1 (10%) | | | |
| Leukemia mononuclear | | 1 (10%) | 1 (10%) | |
| Mesothelioma malignant | | | 3 (30%) | |
| ***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 A1
**Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** | | | | |
| **Alimentary System** | | | | |
| Intestine large, colon | (44) | (48) | (49) | (48) |
| Adenocarcinoma | | | 1 (2%) | |
| Carcinoid tumor malignant | | | | 1 (2%) |
| Intestine large, rectum | (49) | (50) | (50) | (50) |
| Leiomyosarcoma | 1 (2%) | | | |
| Intestine large, cecum | (47) | (50) | (50) | (50) |
| Lipoma | | 1 (2%) | | |
| Intestine small, duodenum | (50) | (50) | (50) | (50) |
| Intestine small, jejunum | (49) | (50) | (50) | (50) |
| Leiomyoma | 1 (2%) | | | |
| Intestine small, ileum | (49) | (50) | (50) | (50) |
| Leiomyosarcoma | | 1 (2%) | | |
| Liver | (50) | (50) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| Carcinoma, metastatic, pancreas | | | | 1 (2%) |
| Hepatocellular carcinoma | | | 1 (2%) | |
| Hepatocellular adenoma | 2 (4%) | 1 (2%) | | 4 (8%) |
| Hepatocellular adenoma, multiple | | 1 (2%) | | |
| Mesentery | (7) | (13) | (12) | (5) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (8%) | |
| Oral mucosa | | (1) | (2) | |
| Squamous cell papilloma | | 1 (100%) | 2 (100%) | |
| Pancreas | (50) | (49) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| Acinus, adenoma | 3 (6%) | 2 (4%) | 3 (6%) | 10 (20%) |
| Acinus, carcinoma | | | | 1 (2%) |
| Salivary glands | (50) | (50) | (50) | (50) |
| Schwannoma malignant | | | | 1 (2%) |
| Stomach, forestomach | (50) | (50) | (50) | (50) |
| Stomach, glandular | (50) | (50) | (50) | (50) |
| Carcinoma | | | | 1 (2%) |
| Tongue | | (1) | (1) | (1) |
| Squamous cell papilloma | | | 1 (100%) | |
| **Cardiovascular System** | | | | |
| Heart | (50) | (50) | (50) | (50) |
| Thymoma malignant, metastatic, thymus | | | | 1 (2%) |

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| *2-Year Study* (continued) | | | | |
| **Endocrine System** | | | | |
| Adrenal cortex | (50) | (50) | (50) | (50) |
| Adrenal medulla | (50) | (50) | (50) | (50) |
| Pheochromocytoma malignant | 2 (4%) | | 2 (4%) | 2 (4%) |
| Pheochromocytoma benign | 6 (12%) | 8 (16%) | 9 (18%) | 8 (16%) |
| Pheochromocytoma benign, multiple | 3 (6%) | 2 (4%) | | |
| Islets, pancreatic | (50) | (49) | (50) | (50) |
| Adenoma | 5 (10%) | 2 (4%) | 4 (8%) | 1 (2%) |
| Pituitary gland | (50) | (50) | (50) | (49) |
| Pars distalis, adenoma | 9 (18%) | 11 (22%) | 11 (22%) | 10 (20%) |
| Pars distalis, adenoma, multiple | 1 (2%) | | | |
| Pars distalis, carcinoma | | 1 (2%) | | |
| Pars intermedia, adenoma | | | 1 (2%) | |
| Thyroid gland | (50) | (50) | (50) | (50) |
| C-cell, adenoma | 5 (10%) | 3 (6%) | 3 (6%) | 1 (2%) |
| Follicular cell, adenoma | | 1 (2%) | | 1 (2%) |
| Follicular cell, carcinoma | | | | 2 (4%) |
| **General Body System** | | | | |
| None | | | | |
| **Genital System** | | | | |
| Epididymis | (50) | (50) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| Preputial gland | (50) | (50) | (50) | (50) |
| Adenoma | 4 (8%) | 3 (6%) | 1 (2%) | |
| Carcinoma | 1 (2%) | 6 (12%) | 1 (2%) | |
| Prostate | (50) | (50) | (50) | (50) |
| Seminal vesicle | (50) | (50) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| Testes | (50) | (50) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| Bilateral, interstitial cell, adenoma | 37 (74%) | 36 (72%) | 40 (80%) | 37 (74%) |
| Interstitial cell, adenoma | 7 (14%) | 10 (20%) | 8 (16%) | 8 (16%) |
| **Hematopoietic System** | | | | |
| Bone marrow | (50) | (50) | (50) | (50) |
| Pheochromocytoma malignant, metastatic, adrenal medulla | | | 1 (2%) | |
| Lymph node | (23) | (22) | (30) | (19) |
| Iliac, leiomyosarcoma, metastatic, intestine large, rectum | 1 (4%) | | | |
| Lymph node, mandibular | (49) | (49) | (50) | (50) |
| Lymph node, mesenteric | (50) | (50) | (50) | (50) |
| Lymph node, mediastinal | | | (1) | |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (100%) | |

TABLE A1
**Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Hematopoietic System** (continued) | | | | |
| Spleen | (50) | (50) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| Fibroma | | 1 (2%) | | |
| Liposarcoma | | 1 (2%) | | |
| Thymus | (49) | (48) | (50) | (48) |
| Thymoma benign | | 1 (2%) | | 1 (2%) |
| Thymoma malignant | | | | 1 (2%) |
| **Integumentary System** | | | | |
| Mammary gland | (45) | (49) | (47) | (48) |
| Carcinoma | | 1 (2%) | | |
| Fibroadenoma | 2 (4%) | 5 (10%) | 2 (4%) | |
| Skin | (50) | (50) | (50) | (50) |
| Basal cell adenoma | | | | 2 (4%) |
| Basal cell carcinoma | 1 (2%) | | 1 (2%) | 1 (2%) |
| Keratoacanthoma | 2 (4%) | 2 (4%) | 2 (4%) | 1 (2%) |
| Squamous cell papilloma | 1 (2%) | | 1 (2%) | 1 (2%) |
| Sebaceous gland, adenoma | | | 1 (2%) | |
| Subcutaneous tissue, fibroma | 5 (10%) | 2 (4%) | 5 (10%) | 5 (10%) |
| Subcutaneous tissue, lipoma | | | | 1 (2%) |
| Subcutaneous tissue, melanoma malignant | 1 (2%) | | | |
| Subcutaneous tissue, schwannoma benign | | 1 (2%) | | |
| Subcutaneous tissue, schwannoma malignant | | 1 (2%) | | 1 (2%) |
| **Musculoskeletal System** | | | | |
| Bone | (50) | (50) | (50) | (50) |
| Chordoma | 1 (2%) | | | |
| Osteosarcoma | | | | 1 (2%) |
| Skeletal muscle | (1) | | (1) | |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (100%) | |
| Hemangiosarcoma | 1 (100%) | | | |
| **Nervous System** | | | | |
| Brain | (50) | (50) | (50) | (50) |
| Astrocytoma malignant | | 1 (2%) | 1 (2%) | |
| Oligodendroglioma malignant | 1 (2%) | | | |
| Spinal cord | (2) | (1) | | (2) |
| **Respiratory System** | | | | |
| Lung | (50) | (50) | (50) | (50) |
| Alveolar/bronchiolar adenoma | | 2 (4%) | | |
| Alveolar/bronchiolar carcinoma | | 1 (2%) | | 1 (2%) |
| Chordoma, metastatic, bone | 1 (2%) | | | |
| Pheochromocytoma malignant, metastatic, adrenal medulla | | | 1 (2%) | |
| Thymoma malignant, metastatic, thymus | | | | 1 (2%) |

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| *2-Year Study* (continued) | | | | |
| **Special Senses System** | | | | |
| None | | | | |
| **Urinary System** | | | | |
| Kidney | (50) | (50) | (50) | (50) |
| Renal tubule, adenoma | 1 (2%) | 2 (4%) | | |
| Renal tubule, carcinoma | | | | 1 (2%) |
| Urinary bladder | (50) | (50) | (50) | (50) |
| Adenocarcinoma, metastatic, intestine large, colon | | | 1 (2%) | |
| **Systemic Lesions** | | | | |
| Multiple organs | (50) | (50) | (50) | (50) |
| Leukemia mononuclear | 31 (62%) | 28 (56%) | 34 (68%) | 30 (60%) |
| Mesothelioma malignant | 1 (2%) | 3 (6%) | | 1 (2%) |
| **Neoplasm Summary** | | | | |
| Total animals with primary neoplasms[c] | | | | |
| 15-Month interim evaluation | 8 | 7 | 9 | 9 |
| 2-Year study | 50 | 50 | 50 | 50 |
| Total primary neoplasms | | | | |
| 15-Month interim evaluation | 12 | 8 | 14 | 11 |
| 2-Year study | 135 | 142 | 135 | 136 |
| Total animals with benign neoplasms | | | | |
| 15-Month interim evaluation | 8 | 7 | 9 | 9 |
| 2-Year study | 46 | 49 | 49 | 47 |
| Total benign neoplasms | | | | |
| 15-Month interim evaluation | 11 | 7 | 10 | 11 |
| 2-Year study | 94 | 98 | 94 | 91 |
| Total animals with malignant neoplasms | | | | |
| 15-Month interim evaluation | 1 | 1 | 4 | |
| 2-Year study | 37 | 35 | 37 | 38 |
| Total malignant neoplasms | | | | |
| 15-Month interim evaluation | 1 | 1 | 4 | |
| 2-Year study | 41 | 44 | 41 | 45 |
| Total animals with metastatic neoplasms | | | | |
| 2-Year study | 2 | | 2 | 2 |
| Total metastatic neoplasms | | | | |
| 2-Year study | 2 | | 12 | 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 Butyl Benzyl Phthalate: 0 ppm**

| Number of Days on Study | 2 5 5 | 2 6 1 | 4 3 8 | 4 7 7 | 4 7 9 | 5 4 1 | 5 4 4 | 5 5 7 | 5 6 2 | 5 7 6 | 5 9 3 | 6 0 7 | 6 1 1 | 6 1 4 | 6 3 1 | 6 3 1 | 6 4 4 | 6 6 2 | 6 7 7 | 7 1 7 | 7 1 8 | 7 1 8 | 7 2 9 | 7 2 9 | 7 2 9 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 0 0 8 | 0 3 9 | 0 5 3 | 0 0 1 | 0 2 5 | 0 1 2 | 0 1 0 | 0 2 2 | 0 4 0 | 0 4 5 | 0 1 8 | 0 0 5 | 0 4 9 | 0 4 2 | 0 0 9 | 0 2 7 | 0 2 3 | 0 4 1 | 0 3 1 | 0 2 1 | 0 1 7 | 0 3 4 | 0 0 4 | 0 0 6 | 0 1 1 |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, colon | + | + | + | + | + | + | M | + | + | M | + | + | + | + | + | M | + | M | + | + | + | + | + | + | + |
| Intestine large, rectum | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Leiomyosarcoma | X | | | | | | | | | | | | | | | | | | | | | | | | |
| Intestine large, cecum | + | A | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, jejunum | + | A | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Leiomyoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Intestine small, ileum | + | A | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Hepatocellular adenoma | | | | | | | | | | | | | | | | | | X | | | | | | | |
| Mesentery | | | | | | | + | | | + | + | | | | | | | | + | | | | | | |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Acinus, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pheochromocytoma malignant | | | | | | | | | | | | | | X | | | | | | | | | | | |
| Pheochromocytoma benign | | | | | | | | | | | | | | | | | X | | | | | | | | |
| Pheochromocytoma benign, multiple | | | | | | | | | | | | X | | | | | | | | | | X | X | | |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pars distalis, adenoma | | | X | | | | X | | | | | | | | | | | | | | X | | | | |
| Pars distalis, adenoma, multiple | | | | | | | | | | | | | | | | | | | | | | | | | |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| C-cell, adenoma | | | X | | | | | | | X | | | | | | | | | X | | | | | | |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Tissue NOS | | | | | | | | | | | | | | | | | + | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | X | | | X | | | | | |
| 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 |
| Interstitial cell, adenoma | | | | | | | X | X | | | | | | | X | | | | | | 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 Butyl Benzyl Phthalate: 0 ppm (continued)

| Number of Days on Study | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 2 9 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 0 1 4 | 0 1 5 | 0 1 9 | 0 2 0 | 0 2 2 | 0 2 4 | 0 2 6 | 0 2 9 | 0 3 3 | 0 3 6 | 0 3 7 | 0 3 8 | 0 4 3 | 0 4 4 | 0 4 6 | 0 4 6 | 0 0 2 | 0 0 7 | 0 1 3 | 0 1 6 | 0 2 3 | 0 2 8 | 0 3 0 | 0 3 2 | 0 3 5 | 0 4 7 | 0 4 8 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | M | + | + | + | 44 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Leiomyosarcoma | | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | 47 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Leiomyoma | X | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Hepatocellular adenoma | | | | | | | X | | | | | | | | | | | | | | | | | | | | 2 |
| Mesentery | | | + | | | | | | | | | | | | | | + | | | | | | | + | | | 7 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Acinus, adenoma | | | X | | | | | | | | X | | | | | | | | | | | X | | | | | 3 |
| 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 | | | | | | | | | | | | | | | | | | X | | | | | | | | | 2 |
| Pheochromocytoma benign | | | | | | | | | | | | | | | | | X | X | X | X | X | | | | | | 6 |
| Pheochromocytoma benign, multiple | | | | | | | | | | | | | | | | | | | | | | | | | | | 3 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | X | | X | | | | | | X | | X | | | | X | | | | | 5 |
| Parathyroid gland | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pars distalis, adenoma | X | | | X | | | | | X | | X | | X | | | | | | X | | | | | | | | 9 |
| Pars distalis, adenoma, multiple | | | | | | | | | | | | | | | | | | | | | | X | | | | | 1 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | | | | | | | X | | | | | | | X | | | | | | | | | 5 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Tissue NOS | | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | X | | | | | | | | | X | | | | | | | | | | | | | | | 4 |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Prostate | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Seminal vesicle | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Testes | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| 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 | 37 |
| Interstitial cell, adenoma | X | | | X | | | | | | | | | | | | | | | | | | X | | | | | 7 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 0 ppm** (continued)

| Number of Days on Study | 2 5 5 | 2 6 1 | 4 3 8 | 4 7 7 | 4 7 9 | 5 4 1 | 5 4 4 | 5 5 7 | 5 6 2 | 5 7 6 | 5 9 3 | 6 0 7 | 6 1 1 | 6 1 4 | 6 3 1 | 6 3 1 | 6 4 4 | 6 6 2 | 6 7 7 | 7 1 1 | 7 1 8 | 7 1 8 | 7 2 9 | 7 2 9 | 7 2 9 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 0 0 8 | 0 3 9 | 0 5 3 | 0 0 1 | 0 2 5 | 0 1 2 | 0 1 0 | 0 2 2 | 0 4 0 | 0 4 5 | 0 1 8 | 0 0 5 | 0 4 9 | 0 4 2 | 0 0 9 | 0 2 7 | 0 0 3 | 0 4 1 | 0 3 1 | 0 2 1 | 0 1 7 | 0 3 4 | 0 0 4 | 0 0 6 | 0 1 1 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood | | | | | | | | | | | | | | | | | | | | | | | | | + |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node | + | + | | | + | + | | + | + | + | + | + | + | | + | | + | | + | + | + | | | + | |
| Iliac, leiomyosarcoma, metastatic, intestine large, rectum | X | | | | | | | | | | | | | | | | | | | | | | | | |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | M | + | + | + | + | + | + | + | + |
| Fibroadenoma | | | | | | | | | | | | | | | | | | | | | | | | | X |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Basal cell carcinoma | | | | | | | | | | | X | | | | | | | | | | | | | | |
| Keratoacanthoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | X | | | | | | | | X | | | |
| Subcutaneous tissue, melanoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Chordoma | | | | | | | | | | | | | | X | | | | | | | | | | | |
| Skeletal muscle | | | + | | | | | | | | | | | | | | | | | | | | | | |
| Hemangiosarcoma | | | X | | | | | | | | | | | | | | | | | | | | | | |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Oligodendroglioma malignant | | X | | | | | | | | | | | | | | | | | | | | | | | |
| Peripheral nerve | | | | | | | | | | | | | | + | | + | | | | | | | | | |
| Spinal cord | | | | | | | | | | | | | | | | + | | | | | | | | | |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Chordoma, metastatic, bone | | | | | | | | | | | | | | X | | | | | | | | | | | |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ear | | | | | | | | | | | | | | | | | | | | | | | | | |
| Eye | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Renal tubule, adenoma | | | | | | | | | | | | | | | | | | | | | | | | 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 |
| Mesothelioma malignant | | | | | | | | | | | | | | | | | | | X | | | | | | |

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 0 ppm (continued)

| Number of Days on Study | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 729 | 730 | 730 | 730 | 730 | 730 | 730 | 730 | 730 | 730 | 730 | 731 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 01 14 | 01 15 | 01 19 | 02 20 | 02 24 | 02 26 | 02 29 | 03 33 | 03 36 | 03 37 | 03 38 | 04 33 | 04 44 | 04 46 | 02 02 | 02 07 | 03 13 | 01 16 | 02 23 | 02 28 | 03 30 | 03 32 | 03 35 | 04 47 | 04 48 | Total Tissues/ Tumors |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node | | | + | | + | + | + | + | + | | | | | | | | | | | | | | | + | | 23 |
| Iliac, leiomyosarcoma, metastatic, intestine large, rectum | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | M | + | + | + | + | + | + | + | + | + | + | M | + | + | + | M | + | + | + | + | + | + | 45 |
| Fibroadenoma | | | | | | | | | | | | | | | | | | | | | | X | | | | 2 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Basal cell carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Keratoacanthoma | | | | | | | | X | | | | | | | | | | | | | | | | X | | 2 |
| Squamous cell papilloma | | | | | | | | | | X | | | | | | | | | | | | | | | | 1 |
| Subcutaneous tissue, fibroma | | | | | | | X | | | | | X | | | | | | | | | | | X | | | 5 |
| Subcutaneous tissue, melanoma malignant | | | | | | | | | | | | | | | | | | | X | | | | | | | 1 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Chordoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Skeletal muscle | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Hemangiosarcoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Oligodendroglioma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Peripheral nerve | | | | | | + | | | | | | | | | | | | | | | | | | | | 3 |
| Spinal cord | | | | | | + | | | | | | | | | | | | | | | | | | | | 2 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Chordoma, metastatic, bone | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ear | | | | | | | | | | | | | | | | | | + | | | + | | | | | 2 |
| Eye | | | | | | | | | | | | | | | | | | + | | | | | | | | 1 |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Renal tubule, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leukemia mononuclear | | X | | X | X | X | X | | | | X | | | X | X | X | | | | | X | X | X | X | 31 |
| Mesothelioma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 3,000 ppm**

| Number of Days on Study | 3 0 3 | 4 5 0 | 4 7 7 | 4 9 9 | 4 9 9 | 5 1 9 | 5 4 1 | 5 4 4 | 5 5 0 | 5 6 5 | 5 6 9 | 5 8 4 | 5 8 6 | 6 0 1 | 6 0 7 | 6 0 7 | 6 0 7 | 6 1 1 | 6 2 6 | 6 2 7 | 6 4 4 | 6 4 5 | 6 4 9 | 6 7 8 | 6 9 4 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 0 6 3 | 0 6 8 | 0 8 5 | 0 6 4 | 0 7 1 | 0 8 9 | 1 0 6 | 0 9 1 | 1 0 7 | 0 9 3 | 1 0 9 | 0 8 6 | 1 0 8 | 1 7 0 | 0 6 7 | 0 7 9 | 0 8 0 | 1 0 3 | 0 7 3 | 0 7 2 | 1 8 4 | 0 4 4 | 0 8 4 | 0 7 4 | 1 6 1 | 0 0 2 |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lipoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Leiomyosarcoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Hepatocellular adenoma | | | | | | | | | | | | | | | | | | | | | | | | X | |
| Hepatocellular adenoma, multiple | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mesentery | | | | | | + | | | + | | | | | | + | | + | | + | | | | + | | |
| Oral mucosa | | | | | | | | | | | | | | | | | | | | | | | | | |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Pancreas | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Acinus, adenoma | | | | | | | | | | | X | | | | | | | | | | | | | | |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Tongue | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pheochromocytoma benign | | | | | | | | X | | | | | | X | | | | | | | | | | | |
| Pheochromocytoma benign, multiple | | | | | | | | | | | | | | | | | | | | | | | | | |
| Islets, pancreatic | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | X | | | | | | | | | | |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pars distalis, adenoma | | | | | X | | | | | | | | X | | | | | | | | X | X | | | |
| Pars distalis, carcinoma | X | | | | | | | | | | | | | | | | | | | | | | | | |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | X | | | | | | |
| Follicular cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | |
| **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 | | |
| Interstitial cell, adenoma | | X | X | | | | X | | X | | | X | X | | | X | | | | | | | | X | X |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 3,000 ppm**
(continued)

| Number of Days on Study | 6 9 8 | 7 0 8 | 7 0 8 | 7 2 3 | 7 2 3 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 0 8 0 | 0 9 0 | 1 0 5 | 0 7 5 | 0 9 2 | 0 6 2 | 0 6 6 | 0 6 7 | 0 7 9 | 0 8 2 | 0 8 7 | 0 9 5 | 0 9 6 | 0 9 7 | 0 9 8 | 0 9 9 | 1 0 8 | 0 6 5 | 0 7 3 | 0 7 6 | 0 8 1 | 0 8 8 | 0 9 4 | 0 9 9 | 1 0 1 | 1 0 9 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, colon | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lipoma | | | | | X | | | | | | | | | | | | | | | | | | | | 1 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leiomyosarcoma | | | | | | | | | | | | | | | | | | | | | X | | | | 1 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Hepatocellular adenoma | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Hepatocellular adenoma, multiple | | | | | | | | | | | | | | | | | | | | | | | | X | 1 |
| Mesentery | | + | | + | + | | + | | | | | | + | | | | | | | + | | | | + | 13 |
| Oral mucosa | | | | | | | | | | | | | | | | | | | | | | + | | | 1 |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | X | | | 1 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Acinus, adenoma | | | | | | | | | | | | | | | | | | | | | | X | | | 2 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Tongue | | | | | | | | | | | | | | + | | | | | | | | | | | 1 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pheochromocytoma benign | | X | | | | | | X | | | | | | X | X | | | | X | X | | | | | 8 |
| Pheochromocytoma benign, multiple | X | | | | | | | | | | | | | | | | | | | | X | | | | 2 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Adenoma | | X | | | | | | | | | | | | | | | | | | | | | | | 2 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pars distalis, adenoma | | | | | | | | X | | X | | | | | | X | X | | X | | | | X | X | 11 |
| Pars distalis, carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | | | | | | X | | | | | | | | | | | | | | X | 3 |
| Follicular cell, adenoma | | X | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | X | | | | | | | | | | X | | | X | | | 3 |
| Carcinoma | | | | X | X | | | | X | | | | | | X | | | | | | | | X | | 6 |
| Prostate | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Seminal vesicle | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Testes | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| 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 | 36 |
| Interstitial cell, adenoma | | | | | | | | | | | | | | | | | | | | X | | | | | 10 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 3,000 ppm**
(continued)

| Number of Days on Study | 3 0 3 | 4 5 0 | 4 7 7 | 4 9 9 | 4 9 9 | 5 1 9 | 5 4 1 | 5 4 4 | 5 5 0 | 5 6 5 | 5 6 9 | 5 8 4 | 5 8 6 | 6 0 1 | 6 0 7 | 6 0 7 | 6 0 7 | 6 1 1 | 6 2 6 | 6 2 7 | 6 4 4 | 6 4 5 | 6 4 9 | 6 7 8 | 6 9 4 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 0 6 3 | 0 6 8 | 0 8 5 | 0 6 4 | 0 7 1 | 0 8 9 | 1 0 6 | 0 9 1 | 1 0 7 | 0 9 3 | 1 0 0 | 0 8 6 | 1 1 0 | 0 7 7 | 0 6 9 | 0 7 0 | 0 8 3 | 0 7 3 | 0 7 2 | 0 7 8 | 1 0 4 | 0 8 4 | 0 7 4 | 0 6 1 | 1 0 2 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node | | + | + | | | | | | + | + | | + | + | | + | + | + | + | | + | + | | | |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Fibroma | | | | | | X | | | | | | | | | | | | | | | | | | | |
| Liposarcoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Thymus | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymoma benign | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Fibroadenoma | | | | | | | | | | | | | | | | | | | | | | X | | X | |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Keratoacanthoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | | | | | X | | | |
| Subcutaneous tissue, schwannoma benign | | | | | | | | X | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, schwannoma malignant | | | | | | | | | | | | | | | | | | | | | | X | | | |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Astrocytoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| Peripheral nerve | | | | | | | | | | | | | | | | | | | | | | | | | + |
| Spinal cord | | | | | | | | | | | | | | | | | | | | | | | | | + |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Alveolar/bronchiolar adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Alveolar/bronchiolar carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Renal tubule, adenoma | | | | | | | | | | | | | | | X | | | | | | | | | | |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Leukemia mononuclear | | | | X | | 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 Butyl Benzyl Phthalate: 3,000 ppm
(continued)

| Number of Days on Study | 6 9 8 | 7 0 8 | 7 0 8 | 7 2 3 | 7 2 3 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 0 8 0 | 0 9 0 | 1 0 5 | 0 7 5 | 0 9 2 | 0 6 2 | 0 6 6 | 0 6 7 | 0 7 9 | 0 8 2 | 0 8 7 | 0 8 9 | 0 9 5 | 0 9 6 | 0 9 7 | 0 9 8 | 0 1 8 | 0 6 5 | 0 7 3 | 0 7 6 | 0 8 1 | 0 8 8 | 0 9 4 | 0 9 9 | 1 0 1 | 1 0 9 | Total Tissues/ Tumors |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node | | + | + | + | | | | + | | + | | + | + | + | | | | | | | + | | + | | | 22 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | 49 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Fibroma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Liposarcoma | | | | | | X | | | | | | | | | | | | | | | | | | | | 1 |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | 48 |
| Thymoma benign | | | | | | | | | | | | | | X | | | | | | | | | | | | 1 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Carcinoma | | | | | | | | | | X | | | | | | | | | | | | | | | | 1 |
| Fibroadenoma | | | X | | | | | | | | | | X | | | | | X | | | | | | | | 5 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Keratoacanthoma | | | | | | X | | | | | | | | | | X | | | | | | | | | | 2 |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | | | | | | | X | | 2 |
| Subcutaneous tissue, schwannoma benign | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Subcutaneous tissue, schwannoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Astrocytoma malignant | | | | | | | | | | | | | | | | | | | | | | | | X | | 1 |
| Peripheral nerve | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Spinal cord | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Alveolar/bronchiolar adenoma | | | | | | X | | | | | | | | | | | | | | X | | | | | | 2 |
| Alveolar/bronchiolar carcinoma | | | | | | | | | | | | | | | | | | | | | | | X | | | 1 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Renal tubule, adenoma | | | | | | | | | | | | | | | | | | | | | | | | X | | 2 |
| 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 | 28 |
| Mesothelioma malignant | | | | | | | | | | | | | | X | | | | | | | | | | | | 3 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm**

| Number of Days on Study | 4 4 5 5 5 5 5 6 6 6 6 6 6 6 6 6 6 6 6 7 7 7 7 7<br>0 9 4 4 6 7 9 1 1 2 5 5 7 7 8 8 8 8 9 9 0 0 0 0 0<br>0 4 2 2 9 4 2 1 4 8 1 5 0 4 1 1 3 4 7 7 1 2 2 3 8 |
|---|---|
| **Carcass ID Number** | 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1<br>3 2 5 6 6 2 7 6 6 4 4 5 4 3 3 5 3 3 2 2 5 3 4 5 6<br>0 9 9 9 0 7 0 8 4 5 4 7 7 5 3 0 7 6 3 8 4 2 9 6 2 |
| **Alimentary System** | |
| Esophagus | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine large, colon | + + + + + + + + + + + + + + M + + + + + + + + + |
| Adenocarcinoma | X |
| Intestine large, rectum | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine large, cecum | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine small, duodenum | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine small, jejunum | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine small, ileum | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Liver | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenocarcinoma, metastatic, intestine large, colon | X |
| Hepatocellular carcinoma | |
| Mesentery | + + + + + + + + + + + + + + + + + + + + + |
| Adenocarcinoma, metastatic, intestine large, colon | X |
| Oral mucosa | |
| Squamous cell papilloma | |
| Pancreas | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenocarcinoma, metastatic, intestine large, colon | X |
| Acinus, adenoma | |
| Salivary glands | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Stomach, forestomach | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Stomach, glandular | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Tongue | + |
| Squamous cell papilloma | X |
| **Cardiovascular System** | |
| Blood vessel | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Heart | + + + + + + + + + + + + + + + + + + + + + + + + + |
| **Endocrine System** | |
| Adrenal cortex | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Adrenal medulla | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Pheochromocytoma malignant | X X |
| Pheochromocytoma benign | X X X |
| Islets, pancreatic | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenoma | X X |
| Parathyroid gland | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Pituitary gland | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Pars distalis, adenoma | X X X |
| Pars intermedia, adenoma | X |
| Thyroid gland | + + + + + + + + + + + + + + + + + + + + + + + + + |
| C-cell, adenoma | X X |
| **General Body System** | |
| None | |

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm
(continued)

| Number of Days on Study | 7 1 7 | 7 2 2 | 7 2 3 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 1 6 5 | 1 2 2 | 1 4 2 | 1 2 4 | 1 2 6 | 1 3 8 | 1 4 1 | 1 4 6 | 1 5 1 | 1 5 3 | 1 5 8 | 1 6 3 | 1 6 7 | 1 2 1 | 1 2 5 | 1 3 1 | 1 3 4 | 1 3 9 | 1 4 0 | 1 4 3 | 1 4 8 | 1 5 2 | 1 5 5 | 1 6 1 | 1 6 6 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Adenocarcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Hepatocellular carcinoma | | X | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Mesentery | | | + | | | + | | | + | | | | | | | | | | | | | | | | | 12 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Oral mucosa | | | | | | | | | | | | | | + | | | | | | | | | | | + | 2 |
| Squamous cell papilloma | | | | | | | | | | | | | | X | | | | | | | | | | | X | 2 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Acinus, adenoma | | | | | | | | | | | | | X | X | | | | | | | | | X | | | 3 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Tongue | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pheochromocytoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| Pheochromocytoma benign | | | | X | | | | | | | X | | X | | | X | | | | | | | X | | X | 9 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | X | X | | 4 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | 49 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pars distalis, adenoma | | | X | | X | | | | | | X | | | | X | | X | | | | | | X | X | | 11 |
| Pars intermedia, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | X | | | | | | | | | | | | | | | | | | | | | 3 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm**
(continued)

| Number of Days on Study | 4 0 0 | 4 9 4 | 5 4 2 | 5 4 2 | 5 6 9 | 5 7 4 | 5 9 2 | 6 1 1 | 6 1 4 | 6 2 8 | 6 5 1 | 6 5 5 | 6 7 0 | 6 7 4 | 6 8 1 | 6 8 1 | 6 8 3 | 6 8 4 | 6 9 7 | 6 9 7 | 7 0 1 | 7 0 2 | 7 0 2 | 7 0 3 | 7 0 8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 1 3 0 | 1 2 9 | 1 5 9 | 1 6 9 | 1 6 0 | 1 2 7 | 1 7 0 | 1 6 8 | 1 6 4 | 1 4 5 | 1 4 4 | 1 5 7 | 1 4 7 | 1 3 5 | 1 3 3 | 1 5 0 | 1 3 7 | 1 3 6 | 1 2 3 | 1 2 8 | 1 5 4 | 1 3 2 | 1 4 9 | 1 5 6 | 1 6 2 |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Prostate | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Seminal vesicle | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| Testes | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| 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 | | | | | X | | | | | | | | | | X |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pheochromocytoma malignant, metastatic, adrenal medulla | | | | | | | | | | | | | | | X | | | | | | | | | | |
| Lymph node | + | | + | + | | + | | + | + | + | + | + | + | + | + | + | + | + | + | | | + | | + | + |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mediastinal | | | | | | | | | | + | | | | | | | | | | | | | | | |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | I | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Fibroadenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Basal cell carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Keratoacanthoma | | | | | | | | | | | | | X | | | | | | | | | | | | |
| Squamous cell papilloma | | | | | | | | | | | | X | | | | | | | | | | | | | |
| Sebaceous gland, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, fibroma | | | | | | | | | | X | | | | X | | | | | | | | X | | | X |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Skeletal muscle | | | | | | | | | | + | | | | | | | | | | | | | | | |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Astrocytoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm
(continued)

| Number of Days on Study | 7 1 7 | 7 2 2 | 7 2 3 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 1 6 5 | 1 2 2 | 1 4 2 | 1 2 4 | 1 2 6 | 1 3 8 | 1 4 1 | 1 4 6 | 1 5 1 | 1 5 3 | 1 5 8 | 1 6 3 | 1 6 7 | 1 2 1 | 1 2 5 | 1 3 1 | 1 3 4 | 1 3 9 | 1 4 0 | 1 4 3 | 1 4 8 | 1 5 2 | 1 5 5 | 1 6 1 | 1 6 6 | Total Tissues/ Tumors |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | | | | | | | | | X | | | | | | 1 |
| Carcinoma | | | | | | | | | | | | X | | | | | | | | | | | | | | 1 |
| Prostate | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Seminal vesicle | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Testes | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| 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 | 40 |
| Interstitial cell, adenoma | | | X | X | | | | | | | | | | | | X | | | | | | | | | | 8 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pheochromocytoma malignant, metastatic, adrenal medulla | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Lymph node | + | + | + | + | | | + | | | | + | | | | | | | + | + | + | | + | + | | | 30 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node, mediastinal | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | 47 |
| Fibroadenoma | | | | | | | | | | | | | | | | | | | X | X | | | | | | 2 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Basal cell carcinoma | | | | X | | | | | | | | | | | | | | | | | | | | | | 1 |
| Keratoacanthoma | | | | | | | | | | | | | | | | | | | | | | | | X | | 2 |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Sebaceous gland, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | X | 1 |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | | | | | | | X | | 5 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Skeletal muscle | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Astrocytoma malignant | | X | | | | | | | | | | | | | | | | | | | | | | | | 1 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm**
(continued)

| Number of Days on Study | 4 0 0 | 4 9 4 | 5 4 2 | 5 4 2 | 5 6 9 | 5 7 4 | 5 9 2 | 6 1 1 | 6 1 4 | 6 2 8 | 6 5 1 | 6 5 5 | 6 7 0 | 6 7 4 | 6 8 1 | 6 8 1 | 6 8 3 | 6 8 4 | 6 9 7 | 6 9 7 | 7 0 1 | 7 0 2 | 7 0 2 | 7 0 3 | 7 0 8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 1 3 0 | 1 2 0 | 1 5 9 | 1 6 9 | 1 6 0 | 1 2 7 | 1 7 0 | 1 6 8 | 1 6 4 | 1 4 5 | 1 4 4 | 1 5 7 | 1 4 7 | 1 3 5 | 1 3 3 | 1 5 3 | 1 3 0 | 1 3 7 | 1 2 6 | 1 2 3 | 1 5 4 | 1 3 2 | 1 4 9 | 1 5 6 | 1 6 2 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pheochromocytoma malignant, metastatic, adrenal medulla | | | | | | | | | | | | | | | X | | | | | | | | | | |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | X | | | | | | | | | | | | | | | |
| **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 | |

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm
(continued)

| Number of Days on Study | 7 1 7 | 7 2 2 | 7 2 3 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 1 6 5 | 1 2 2 | 1 4 2 | 1 2 4 | 1 2 6 | 1 3 8 | 1 4 1 | 1 4 6 | 1 5 1 | 1 5 3 | 1 5 8 | 1 6 3 | 1 6 7 | 1 2 1 | 1 2 5 | 1 3 1 | 1 3 4 | 1 3 9 | 1 4 0 | 1 4 3 | 1 4 8 | 1 5 2 | 1 5 5 | 1 6 1 | 1 6 6 | Total Tissues/ Tumors |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pheochromocytoma malignant, metastatic, adrenal medulla | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenocarcinoma, metastatic, intestine large, colon | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leukemia mononuclear | X | X | X | X | | | X | X | | X | X | | X | | | | | | X | X | X | X | X | | | 34 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm**

| Number of Days on Study | 1 8 0 | 2 9 7 | 4 6 4 | 4 6 6 | 4 7 7 | 5 5 0 | 5 9 3 | 6 1 4 | 6 3 4 | 6 3 9 | 6 3 9 | 6 5 4 | 6 6 2 | 6 6 3 | 6 6 8 | 6 7 4 | 6 7 4 | 6 7 5 | 6 8 4 | 6 9 8 | 6 9 8 | 7 0 3 | 7 0 9 | 7 0 9 | 7 1 6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 2 1 9 | 1 8 6 | 1 8 7 | 2 0 1 | 2 2 4 | 2 0 7 | 2 1 6 | 2 2 5 | 1 8 4 | 1 8 5 | 2 2 8 | 1 9 3 | 1 8 8 | 2 0 6 | 2 0 2 | 1 8 9 | 2 0 4 | 2 1 5 | 1 9 0 | 1 8 1 | 1 9 4 | 1 9 1 | 2 1 7 | 2 2 7 | 2 0 5 |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoid tumor malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoma, metastatic, pancreas | | | | | | | | | | | | | | | | X | | | | | | | | | |
| Hepatocellular adenoma | | | X | | | | | | | | | | | | | | | | | | | | | X | |
| Mesentery | | | | | | | | | + | | | | + | | | | + | | | | | | | + | + |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Acinus, adenoma | | | | | | | | | | | | | | | | | | | | | | | | X | X |
| Acinus, carcinoma | | | | | | | | | | | | | | | | X | | | | | | | | | |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Schwannoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Tongue | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymoma malignant, metastatic, thymus | | X | | | | | | | | | | | | | | | | | | | | | | | |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pheochromocytoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| Pheochromocytoma benign | | | | | | | | | | | X | | | | X | | X | | X | X | | | | | |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pituitary gland | I | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pars distalis, adenoma | | | | | | | | | | | | | | | | | | | X | X | | X | | X | X |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | X | |
| Follicular cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Follicular cell, carcinoma | | | | | | | | | | | | | | | | | | | | X | | | | | |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Prostate | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Seminal vesicle | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Testes | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Bilateral, interstitial cell, adenoma | | | | | | | X | | X | X | X | | X | X | X | X | X | X | X | X | X | X | X | | |
| Interstitial cell, adenoma | | | X | X | | X | | X | | | | X | | | | | | | | | | | | | |

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm
(continued)

| Number of Days on Study | 7 1 6 | 7 1 6 | 7 1 8 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 2 1 1 | 2 1 2 | 2 0 9 | 1 8 3 | 1 9 5 | 1 9 8 | 2 0 0 | 2 1 0 | 2 1 8 | 2 2 0 | 2 2 2 | 2 2 6 | 2 3 0 | 1 8 2 | 1 9 2 | 1 9 6 | 1 9 7 | 1 9 9 | 2 0 3 | 2 0 8 | 2 1 3 | 2 1 4 | 2 2 1 | 2 2 3 | 2 2 9 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, colon | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | 48 |
| Carcinoid tumor malignant | | | X | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Carcinoma, metastatic, pancreas | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Hepatocellular adenoma | | | | | | | | | | | | | | | | | X | X | | | | | | | | 4 |
| Mesentery | | | | | | | | | | | | | | | | | | | | | | | | | | 5 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Acinus, adenoma | | | | X | | | | | | | X | X | | | | X | | X | | X | | X | | | X | 10 |
| Acinus, carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Schwannoma malignant | | | X | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Carcinoma | | | | | | | | | | | | | X | | | | | | | | | | | | | 1 |
| Tongue | | | | | | | | | | | | | + | | | | | | | | | | | | | 1 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymoma malignant, metastatic, thymus | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pheochromocytoma malignant | | | | | X | | | | | | | | | X | | | | | | | | | | | | 2 |
| Pheochromocytoma benign | | | | | | | | | | | X | | | | | | | | | | | | | X | X | 8 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | X | | | | | | | | | | | | | | | | | | | | 1 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pars distalis, adenoma | | | | | | | | | X | X | | | | | | X | X | | | X | | | | | | 10 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Follicular cell, adenoma | | | X | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Follicular cell, carcinoma | | | | | | | | X | | | | | | | | | | | | | | | | | | 2 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Epididymis | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Preputial gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Prostate | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Seminal vesicle | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Testes | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| 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 | | 37 |
| Interstitial cell, adenoma | | | | | | | | | | | | X | | | X | | | | | | | | | | X | 8 |

TABLE A2
**Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm**
(continued)

| Number of Days on Study | 1 8 0 | 2 9 7 | 4 6 4 | 4 6 6 | 4 7 7 | 5 5 0 | 5 9 3 | 6 1 4 | 6 3 4 | 6 3 9 | 6 3 9 | 6 5 4 | 6 6 2 | 6 6 3 | 6 6 8 | 6 7 4 | 6 7 4 | 6 7 5 | 6 8 4 | 6 9 8 | 6 9 8 | 7 0 3 | 7 0 9 | 7 0 9 | 7 1 6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 2 1 9 | 1 8 6 | 1 8 7 | 2 0 1 | 2 2 4 | 2 0 7 | 2 1 6 | 2 2 5 | 1 8 4 | 1 8 5 | 2 2 8 | 1 9 3 | 1 8 8 | 2 0 6 | 2 0 2 | 1 8 9 | 2 0 4 | 2 1 5 | 1 9 0 | 1 8 1 | 1 8 4 | 1 9 1 | 2 1 7 | 2 2 7 | 2 0 5 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node | + | | | | + | + | + | + | | | + | + | + | + | + | | + | + | | | + | | | + | |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + |
| Thymoma benign | | | | | | | | | | | | | | | | | | | | | | | | | |
| Thymoma malignant | | X | | | | | | | | | | | | | | | | | | | | | | | |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | M | + | + | + | + |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Basal cell adenoma | | | | | | | | | | | X | | | | | | | | | | | | | | |
| Basal cell carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Keratoacanthoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | X | | | | X | | | X | | | |
| Subcutaneous tissue, lipoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, schwannoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Osteosarcoma | | | | X | | | | | | | | | | | | | | | | | | | | | |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Peripheral nerve | | | | + | | | | | | | | | | | | | | | | | | | | | |
| Spinal cord | | | | + | | | | | | | | | | | | | | | | | | | | | + |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Alveolar/bronchiolar carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Thymoma malignant, metastatic, thymus | | X | | | | | | | | | | | | | | | | | | | | | | | |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ear | | | | | | | | | | | | | | | | | | | | | | | | | + |
| Eye | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Renal tubule, carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | 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 | |
| Mesothelioma malignant | | | | | | | | | X | | | | | | | | | | | | | | | | |

TABLE A2
Individual Animal Tumor Pathology of Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm
(continued)

| Number of Days on Study | 7 1 6 | 7 1 6 | 7 1 8 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 0 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | 7 3 1 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 2 1 1 | 2 1 2 | 2 0 9 | 1 8 3 | 1 9 5 | 1 9 8 | 2 0 0 | 2 1 0 | 2 1 8 | 2 2 0 | 2 2 2 | 2 2 6 | 2 3 0 | 1 8 2 | 1 9 2 | 1 9 6 | 1 9 7 | 1 9 9 | 2 0 3 | 2 0 8 | 2 1 3 | 2 1 4 | 2 2 1 | 2 2 3 | 2 2 9 | Total Tissues/ Tumors |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node | + | | | | | | | | | | | | | | | | + | + | + | | + | | | | | 19 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymus | + | + | + | + | + | + | + | I | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Thymoma benign | | | | | | | X | | | | | | | | | | | | | | | | | | | 1 |
| Thymoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Basal cell adenoma | | | | | | | | | | | | | X | | | | | | | | | | | | | 2 |
| Basal cell carcinoma | | | | | | | | | | | | | | | | | X | | | | | | | | | 1 |
| Keratoacanthoma | | | | | | | | | | | | | | | | | | X | | | | | | | | 1 |
| Squamous cell papilloma | | | | | X | | | | | | | | | | | | | | | | | | | | | 1 |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | X | X | | | | | | | 5 |
| Subcutaneous tissue, lipoma | | | | | | | | | | | | X | | | | | | | | | | | | | | 1 |
| Subcutaneous tissue, schwannoma malignant | | | X | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Osteosarcoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Peripheral nerve | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Spinal cord | | | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Alveolar/bronchiolar carcinoma | | | | | | | | | | | | | | | | X | | | | | | | | | | 1 |
| Thymoma malignant, metastatic, thymus | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ear | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Eye | | | | | | + | | | | | | | | | | | | | | | | | | | | 1 |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Renal tubule, carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leukemia mononuclear | X | X | X | | | | | | | | | | X | | | | X | X | X | | X | | X | X | | 30 |
| Mesothelioma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |

TABLE A3
**Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Adrenal Medulla: Benign Pheochromocytoma** | | | | |
| Overall rate[a] | 9/50 (18%) | 10/50 (20%) | 9/50 (18%) | 8/50 (16%) |
| Adjusted rate[b] | 28.2% | 38.8% | 34.5% | 25.4% |
| Terminal rate[c] | 6/28 (21%) | 6/20 (30%) | 6/22 (27%) | 3/22 (14%) |
| First incidence (days) | 607 | 544 | 684 | 639 |
| Life table test[d] | P=0.473N | P=0.263 | P=0.448 | P=0.589N |
| Logistic regression test[d] | P=0.335N | P=0.410 | P=0.562N | P=0.475N |
| Cochran-Armitage test[d] | P=0.402N | | | |
| Fisher exact test[d] | | P=0.500 | P=0.602N | P=0.500N |
| **Adrenal Medulla: Benign or Malignant Pheochromocytoma** | | | | |
| Overall rate | 10/50 (20%) | 10/50 (20%) | 11/50 (22%) | 10/50 (20%) |
| Adjusted rate | 30.1% | 38.8% | 38.6% | 33.3% |
| Terminal rate | 6/28 (21%) | 6/20 (30%) | 6/22 (27%) | 5/22 (23%) |
| First incidence (days) | 607 | 544 | 681 | 639 |
| Life table test | P=0.469 | P=0.348 | P=0.370 | P=0.496 |
| Logistic regression test | P=0.487N | P=0.524 | P=0.557 | P=0.573N |
| Cochran-Armitage test | P=0.535 | | | |
| Fisher exact test | | P=0.598N | P=0.500 | P=0.598N |
| **Liver: Hepatocellular Adenoma** | | | | |
| Overall rate | 2/50 (4%) | 2/50 (4%) | 0/50 (0%) | 4/50 (8%) |
| Adjusted rate | 6.5% | 8.5% | 0.0% | 14.2% |
| Terminal rate | 1/28 (4%) | 1/20 (5%) | 0/22 (0%) | 2/22 (9%) |
| First incidence (days) | 662 | 678 | —[e] | 464 |
| Life table test | P=0.223 | P=0.600 | P=0.250N | P=0.291 |
| Logistic regression test | P=0.240 | P=0.661 | P=0.227N | P=0.339 |
| Cochran-Armitage test | P=0.232 | | | |
| Fisher exact test | | P=0.691N | P=0.247N | P=0.339 |
| **Liver: Hepatocellular Adenoma or Carcinoma** | | | | |
| Overall rate | 2/50 (4%) | 2/50 (4%) | 1/50 (2%) | 4/50 (8%) |
| Adjusted rate | 6.5% | 8.5% | 4.2% | 14.2% |
| Terminal rate | 1/28 (4%) | 1/20 (5%) | 0/22 (0%) | 2/22 (9%) |
| First incidence (days) | 662 | 678 | 722 | 464 |
| Life table test | P=0.212 | P=0.600 | P=0.529N | P=0.291 |
| Logistic regression test | P=0.238 | P=0.661 | P=0.483N | P=0.339 |
| Cochran-Armitage test | P=0.227 | | | |
| Fisher exact test | | P=0.691N | P=0.500N | P=0.339 |
| **Lung: Alveolar/bronchiolar Adenoma or Carcinoma** | | | | |
| Overall rate | 0/50 (0%) | 3/50 (6%) | 0/50 (0%) | 1/50 (2%) |
| Adjusted rate | 0.0% | 15.0% | 0.0% | 4.5% |
| Terminal rate | 0/28 (0%) | 3/20 (15%) | 0/22 (0%) | 1/22 (5%) |
| First incidence (days) | — | 729 (T) | — | 729 (T) |
| Life table test | P=0.581 | P=0.067 | — | P=0.452 |
| Logistic regression test | P=0.581 | P=0.067 | — | P=0.452 |
| Cochran-Armitage test | P=0.634 | | | |
| Fisher exact test | | P=0.121 | — | P=0.500 |

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Mammary Gland: Fibroadenoma** | | | | |
| Overall rate | 2/50 (4%) | 5/50 (10%) | 2/50 (4%) | 0/50 (0%) |
| Adjusted rate | 7.1% | 19.8% | 9.1% | 0.0% |
| Terminal rate | 2/28 (7%) | 2/20 (10%) | 2/22 (9%) | 0/22 (0%) |
| First incidence (days) | 729 (T) | 645 | 729 (T) | — |
| Life table test | P=0.115N | P=0.128 | P=0.607 | P=0.292N |
| Logistic regression test | P=0.081N | P=0.165 | P=0.607 | P=0.292N |
| Cochran-Armitage test | P=0.092N | | | |
| Fisher exact test | | P=0.218 | P=0.691N | P=0.247N |
| **Mammary Gland: Fibroadenoma or Carcinoma** | | | | |
| Overall rate | 2/50 (4%) | 6/50 (12%) | 2/50 (4%) | 0/50 (0%) |
| Adjusted rate | 7.1% | 24.3% | 9.1% | 0.0% |
| Terminal rate | 2/28 (7%) | 3/20 (15%) | 2/22 (9%) | 0/22 (0%) |
| First incidence (days) | 729 (T) | 645 | 729 (T) | — |
| Life table test | P=0.099N | P=0.066 | P=0.607 | P=0.292N |
| Logistic regression test | P=0.066N | P=0.090 | P=0.607 | P=0.292N |
| Cochran-Armitage test | P=0.077N | | | |
| Fisher exact test | | P=0.134 | P=0.691N | P=0.247N |
| **Oral Cavity (Oral Mucosa or Tongue): Squamous Cell Papilloma** | | | | |
| Overall rate | 0/50 (0%) | 1/50 (2%) | 3/50 (6%) | 0/50 (0%) |
| Adjusted rate | 0.0% | 5.0% | 11.9% | 0.0% |
| Terminal rate | 0/28 (0%) | 1/20 (5%) | 2/22 (9%) | 0/22 (0%) |
| First incidence (days) | — | 729 (T) | 697 | — |
| Life table test | P=0.599 | P=0.433 | P=0.097 | — |
| Logistic regression test | P=0.624N | P=0.433 | P=0.121 | — |
| Cochran-Armitage test | P=0.634 | | | |
| Fisher exact test | | P=0.500 | P=0.121 | — |
| **Pancreas: Adenoma** | | | | |
| Overall rate | 3/50 (6%) | 2/49 (4%) | 3/50 (6%) | 10/50 (20%) |
| Adjusted rate | 10.7% | 7.4% | 13.6% | 41.0% |
| Terminal rate | 3/28 (11%) | 1/20 (5%) | 3/22 (14%) | 8/22 (36%) |
| First incidence (days) | 729 (T) | 569 | 729 (T) | 709 |
| Life table test | P=0.002 | P=0.624N | P=0.548 | P=0.011 |
| Logistic regression test | P=0.006 | P=0.543N | P=0.548 | P=0.016 |
| Cochran-Armitage test | P=0.005 | | | |
| Fisher exact test | | P=0.510N | P=0.661N | P=0.036 |
| **Pancreas: Adenoma or Carcinoma** | | | | |
| Overall rate | 3/50 (6%) | 2/49 (4%) | 3/50 (6%) | 11/50 (22%) |
| Adjusted rate | 10.7% | 7.4% | 13.6% | 42.7% |
| Terminal rate | 3/28 (11%) | 1/20 (5%) | 3/22 (14%) | 8/22 (36%) |
| First incidence (days) | 729 (T) | 569 | 729 (T) | 674 |
| Life table test | P<0.001 | P=0.624N | P=0.548 | P=0.007 |
| Logistic regression test | P=0.003 | P=0.543N | P=0.548 | P=0.014 |
| Cochran-Armitage test | P=0.002 | | | |
| Fisher exact test | | P=0.510N | P=0.661N | P=0.020 |

TABLE A3
**Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Pancreatic Islets: Adenoma** | | | | |
| Overall rate | 5/50 (10%) | 2/49 (4%) | 4/50 (8%) | 1/50 (2%) |
| Adjusted rate | 17.9% | 6.8% | 15.2% | 4.5% |
| Terminal rate | 5/28 (18%) | 0/20 (0%) | 2/22 (9%) | 1/22 (5%) |
| First incidence (days) | 729 (T) | 607 | 701 | 729 (T) |
| Life table test | P=0.150N | P=0.347N | P=0.623N | P=0.161N |
| Logistic regression test | P=0.097N | P=0.271N | P=0.518N | P=0.161N |
| Cochran-Armitage test | P=0.112N | | | |
| Fisher exact test | | P=0.226N | P=0.500N | P=0.102N |
| **Pituitary Gland (Pars Distalis): Adenoma** | | | | |
| Overall rate | 10/50 (20%) | 11/50 (22%) | 11/50 (22%) | 10/49 (20%) |
| Adjusted rate | 30.6% | 42.2% | 37.9% | 35.1% |
| Terminal rate | 7/28 (25%) | 7/20 (35%) | 6/22 (27%) | 5/22 (23%) |
| First incidence (days) | 438 | 499 | 569 | 684 |
| Life table test | P=0.473 | P=0.261 | P=0.352 | P=0.431 |
| Logistic regression test | P=0.467N | P=0.460 | P=0.545 | P=0.575N |
| Cochran-Armitage test | P=0.546N | | | |
| Fisher exact test | | P=0.500 | P=0.500 | P=0.579 |
| **Pituitary Gland (Pars Distalis): Adenoma or Carcinoma** | | | | |
| Overall rate | 10/50 (20%) | 12/50 (24%) | 11/50 (22%) | 10/49 (20%) |
| Adjusted rate | 30.6% | 43.3% | 37.9% | 35.1% |
| Terminal rate | 7/28 (25%) | 7/20 (35%) | 6/22 (27%) | 5/22 (23%) |
| First incidence (days) | 438 | 303 | 569 | 684 |
| Life table test | P=0.506 | P=0.197 | P=0.352 | P=0.431 |
| Logistic regression test | P=0.456N | P=0.394 | P=0.545 | P=0.575N |
| Cochran-Armitage test | P=0.511N | | | |
| Fisher exact test | | P=0.405 | P=0.500 | P=0.579 |
| **Preputial Gland: Adenoma** | | | | |
| Overall rate | 4/50 (8%) | 3/50 (6%) | 1/50 (2%) | 0/50 (0%) |
| Adjusted rate | 12.8% | 15.0% | 4.5% | 0.0% |
| Terminal rate | 2/28 (7%) | 3/20 (15%) | 1/22 (5%) | 0/22 (0%) |
| First incidence (days) | 644 | 729 (T) | 729 (T) | — |
| Life table test | P=0.038N | P=0.644 | P=0.231N | P=0.087N |
| Logistic regression test | P=0.022N | P=0.593N | P=0.165N | P=0.059N |
| Cochran-Armitage test | P=0.025N | | | |
| Fisher exact test | | P=0.500N | P=0.181N | P=0.059N |
| **Preputial Gland: Carcinoma** | | | | |
| Overall rate | 1/50 (2%) | 6/50 (12%) | 1/50 (2%) | 0/50 (0%) |
| Adjusted rate | 2.7% | 25.1% | 4.5% | 0.0% |
| Terminal rate | 0/28 (0%) | 3/20 (15%) | 1/22 (5%) | 0/22 (0%) |
| First incidence (days) | 614 | 626 | 729 (T) | — |
| Life table test | P=0.135N | P=0.030 | P=0.749 | P=0.470N |
| Logistic regression test | P=0.099N | P=0.044 | P=0.763N | P=0.504N |
| Cochran-Armitage test | P=0.111N | | | |
| Fisher exact test | | P=0.056 | P=0.753N | P=0.500N |

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Preputial Gland: Adenoma or Carcinoma** | | | | |
| Overall rate | 5/50 (10%) | 8/50 (16%) | 2/50 (4%) | 0/50 (0%) |
| Adjusted rate | 15.1% | 33.9% | 9.1% | 0.0% |
| Terminal rate | 2/28 (7%) | 5/20 (25%) | 2/22 (9%) | 0/22 (0%) |
| First incidence (days) | 614 | 626 | 729 (T) | — |
| Life table test | P=0.014N | P=0.136 | P=0.273N | P=0.045N |
| Logistic regression test | P=0.006N | P=0.201 | P=0.196N | P=0.032N |
| Cochran-Armitage test | P=0.008N | | | |
| Fisher exact test | | P=0.277 | P=0.218N | P=0.028N |
| **Skin: Squamous Cell Papilloma or Keratoacanthoma** | | | | |
| Overall rate | 3/50 (6%) | 2/50 (4%) | 3/50 (6%) | 2/50 (4%) |
| Adjusted rate | 10.7% | 10.0% | 9.4% | 9.1% |
| Terminal rate | 3/28 (11%) | 2/20 (10%) | 1/22 (5%) | 2/22 (9%) |
| First incidence (days) | 729 (T) | 729 (T) | 655 | 729 (T) |
| Life table test | P=0.506N | P=0.654N | P=0.614 | P=0.611N |
| Logistic regression test | P=0.426N | P=0.654N | P=0.641N | P=0.611N |
| Cochran-Armitage test | P=0.456N | | | |
| Fisher exact test | | P=0.500N | P=0.661N | P=0.500N |
| **Skin: Basal Cell Adenoma or Basal Cell Carcinoma** | | | | |
| Overall rate | 1/50 (2%) | 0/50 (0%) | 1/50 (2%) | 3/50 (6%) |
| Adjusted rate | 2.5% | 0.0% | 4.5% | 11.3% |
| Terminal rate | 0/28 (0%) | 0/20 (0%) | 1/22 (5%) | 2/22 (9%) |
| First incidence (days) | 593 | — | 729 (T) | 639 |
| Life table test | P=0.098 | P=0.516N | P=0.742 | P=0.289 |
| Logistic regression test | P=0.101 | P=0.502N | P=0.760 | P=0.307 |
| Cochran-Armitage test | P=0.097 | | | |
| Fisher exact test | | P=0.500N | P=0.753N | P=0.309 |
| **Skin: Squamous Cell Papilloma, Keratoacanthoma, Basal Cell Adenoma, or Basal Cell Carcinoma** | | | | |
| Overall rate | 4/50 (8%) | 2/50 (4%) | 4/50 (8%) | 5/50 (10%) |
| Adjusted rate | 12.9% | 10.0% | 13.8% | 20.2% |
| Terminal rate | 3/28 (11%) | 2/20 (10%) | 2/22 (9%) | 4/22 (18%) |
| First incidence (days) | 593 | 729 (T) | 655 | 639 |
| Life table test | P=0.272 | P=0.478N | P=0.587 | P=0.403 |
| Logistic regression test | P=0.347 | P=0.394N | P=0.618N | P=0.518 |
| Cochran-Armitage test | P=0.309 | | | |
| Fisher exact test | | P=0.339N | P=0.643N | P=0.500 |
| **Skin (Subcutaneous Tissue): Fibroma** | | | | |
| Overall rate | 5/50 (10%) | 2/50 (4%) | 5/50 (10%) | 5/50 (10%) |
| Adjusted rate | 16.0% | 8.3% | 15.9% | 17.3% |
| Terminal rate | 3/28 (11%) | 1/20 (5%) | 1/22 (5%) | 2/22 (9%) |
| First incidence (days) | 614 | 645 | 628 | 668 |
| Life table test | P=0.407 | P=0.343N | P=0.562 | P=0.550 |
| Logistic regression test | P=0.452 | P=0.254N | P=0.601N | P=0.614N |
| Cochran-Armitage test | P=0.415 | | | |
| Fisher exact test | | P=0.218N | P=0.630N | P=0.630N |

TABLE A3
**Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Testes: Adenoma** | | | | |
| Overall rate | 44/50 (88%) | 46/50 (92%) | 48/50 (96%) | 45/50 (90%) |
| Adjusted rate | 97.8% | 97.8% | 98.0% | 100.0% |
| Terminal rate | 27/28 (96%) | 19/20 (95%) | 21/22 (95%) | 22/22 (100%) |
| First incidence (days) | 477 | 450 | 494 | 464 |
| Life table test | P=0.354 | P=0.049 | P=0.122 | P=0.205 |
| Logistic regression test | P=0.568 | P=0.371 | P=0.383 | P=0.619 |
| Cochran-Armitage test | P=0.449 | | | |
| Fisher exact test | | P=0.370 | P=0.134 | P=0.500 |
| **Thyroid Gland (C-cell): Adenoma** | | | | |
| Overall rate | 5/50 (10%) | 3/50 (6%) | 3/50 (6%) | 1/50 (2%) |
| Adjusted rate | 14.1% | 12.8% | 9.7% | 3.6% |
| Terminal rate | 2/28 (7%) | 2/20 (10%) | 1/22 (5%) | 0/22 (0%) |
| First incidence (days) | 438 | 626 | 674 | 709 |
| Life table test | P=0.091N | P=0.475N | P=0.375N | P=0.130N |
| Logistic regression test | P=0.078N | P=0.356N | P=0.378N | P=0.105N |
| Cochran-Armitage test | P=0.079N | | | |
| Fisher exact test | | P=0.357N | P=0.357N | P=0.102N |
| **Thyroid Gland (Follicular Cell): Adenoma or Carcinoma** | | | | |
| Overall rate | 0/50 (0%) | 1/50 (2%) | 0/50 (0%) | 3/50 (6%) |
| Adjusted rate | 0.0% | 4.2% | 0.0% | 11.6% |
| Terminal rate | 0/28 (0%) | 0/20 (0%) | 0/22 (0%) | 1/22 (5%) |
| First incidence (days) | — | 708 | — | 698 |
| Life table test | P=0.042 | P=0.449 | — | P=0.096 |
| Logistic regression test | P=0.050 | P=0.473 | — | P=0.121 |
| Cochran-Armitage test | P=0.044 | | | |
| Fisher exact test | | P=0.500 | — | P=0.121 |
| **All Organs: Malignant Mesothelioma** | | | | |
| Overall rate | 1/50 (2%) | 3/50 (6%) | 0/50 (0%) | 1/50 (2%) |
| Adjusted rate | 3.1% | 10.9% | 0.0% | 2.4% |
| Terminal rate | 0/28 (0%) | 1/20 (5%) | 0/22 (0%) | 0/22 (0%) |
| First incidence (days) | 677 | 607 | — | 634 |
| Life table test | P=0.376N | P=0.245 | P=0.477N | P=0.737N |
| Logistic regression test | P=0.411N | P=0.299 | P=0.497N | P=0.762 |
| Cochran-Armitage test | P=0.409N | | | |
| Fisher exact test | | P=0.309 | P=0.500N | P=0.753N |
| **All Organs: Mononuclear Cell Leukemia** | | | | |
| Overall rate | 31/50 (62%) | 28/50 (56%) | 34/50 (68%) | 30/50 (60%) |
| Adjusted rate | 71.8% | 72.0% | 74.1% | 65.6% |
| Terminal rate | 16/28 (57%) | 10/20 (50%) | 11/22 (50%) | 7/22 (32%) |
| First incidence (days) | 479 | 499 | 400 | 180 |
| Life table test | P=0.469 | P=0.364 | P=0.268 | P=0.478 |
| Logistic regression test | P=0.466 | P=0.233N | P=0.469 | P=0.492N |
| Cochran-Armitage test | P=0.510 | | | |
| Fisher exact test | | P=0.342N | P=0.338 | P=0.500N |

TABLE A3
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **All Organs: Benign Neoplasms** | | | | |
| Overall rate | 46/50 (92%) | 49/50 (98%) | 49/50 (98%) | 47/50 (94%) |
| Adjusted rate | 100.0% | 100.0% | 100.0% | 100.0% |
| Terminal rate | 28/28 (100%) | 20/20 (100%) | 22/22 (100%) | 22/22 (100%) |
| First incidence (days) | 438 | 450 | 494 | 464 |
| Life table test | P=0.374 | P=0.033 | P=0.144 | P=0.199 |
| Logistic regression test | P=0.590 | P=0.204 | P=0.583 | P=0.601 |
| Cochran-Armitage test | P=0.523 | | | |
| Fisher exact test | | P=0.181 | P=0.181 | P=0.500 |
| **All Organs: Malignant Neoplasms** | | | | |
| Overall rate | 37/50 (74%) | 35/50 (70%) | 37/50 (74%) | 38/50 (76%) |
| Adjusted rate | 78.5% | 80.8% | 77.8% | 77.5% |
| Terminal rate | 18/28 (64%) | 12/20 (60%) | 12/22 (55%) | 11/22 (50%) |
| First incidence (days) | 255 | 303 | 400 | 180 |
| Life table test | P=0.394 | P=0.279 | P=0.414 | P=0.342 |
| Logistic regression test | P=0.331 | P=0.396N | P=0.526 | P=0.469 |
| Cochran-Armitage test | P=0.383 | | | |
| Fisher exact test | | P=0.412N | P=0.590N | P=0.500 |
| **All Organs: Benign or Malignant Neoplasms** | | | | |
| Overall rate | 50/50 (100%) | 50/50 (100%) | 50/50 (100%) | 50/50 (100%) |
| Adjusted rate | 100.0% | 100.0% | 100.0% | 100.0% |
| Terminal rate | 28/28 (100%) | 20/20 (100%) | 22/22 (100%) | 22/22 (100%) |
| First incidence (days) | 255 | 303 | 400 | 180 |
| Life table test | P=0.397 | P=0.093 | P=0.285 | P=0.260 |
| Logistic regression test | —[f] | — | — | — |
| Cochran-Armitage test | — | | | |
| Fisher exact test | | P=1.000N | P=1.000N | P=1.000N |

(T)Terminal sacrifice
[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal gland, 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 Pancreatic Neoplasms in Untreated Male F344/N Rats[a]**

| | Incidence in Controls | | |
|---|---|---|---|
| | Adenoma | Carcinoma | Adenoma or Carcinoma |
| **Overall Historical Incidence** | | | |
| Total | 19/1,191 (1.6%) | 0/1,191 (0.0%) | 19/1,191 (1.6%) |
| Standard deviation | 2.4% | | 2.4% |
| Range | 0%-10% | | 0%-10% |

[a] Data as of 17 June 1994

TABLE A4b
**Historical Incidence of Preputial Gland Neoplasms in Untreated Male F344/N Rats[a]**

| | Incidence in Controls | | |
|---|---|---|---|
| | Adenoma | Carcinoma | Adenoma or Carcinoma |
| **Overall Historical Incidence** | | | |
| Total | 84/1,164 (7.2%) | 41/1,164 (3.5%) | 124/1,164 (10.7%) |
| Standard deviation | 5.6% | 3.4% | 7.5% |
| Range | 0%-24% | 0%-14% | 2%-30% |

[a] Data as of 17 June 1994

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Disposition Summary** | | | | |
| Animals initially in study | 60 | 60 | 60 | 60 |
| *15-Month interim evaluation* | 10 | 10 | 10 | 10 |
| Early deaths | | | | |
| Moribund | 19 | 19 | 26 | 26 |
| Natural deaths | 3 | 11 | 2 | 2 |
| Survivors | | | | |
| Terminal sacrifice | 28 | 20 | 22 | 22 |
| Animals examined microscopically | 60 | 60 | 60 | 60 |
| ***15-Month Interim Evaluation*** | | | | |
| **Alimentary System** | | | | |
| Intestine large, colon | (10) | (10) | (10) | (10) |
| Parasite metazoan | 1 (10%) | | 1 (10%) | |
| Intestine large, rectum | (10) | (10) | (10) | (9) |
| Parasite metazoan | 2 (20%) | | 2 (20%) | 1 (11%) |
| Intestine large, cecum | (10) | (10) | (10) | (10) |
| Edema | 1 (10%) | | | |
| Parasite metazoan | | 1 (10%) | | |
| Liver | (10) | (10) | (10) | (10) |
| Angiectasis | 1 (10%) | | | |
| Basophilic focus | 6 (60%) | 4 (40%) | 2 (20%) | 1 (10%) |
| Clear cell focus | 2 (20%) | 2 (20%) | 1 (10%) | 1 (10%) |
| Eosinophilic focus | 3 (30%) | 1 (10%) | 1 (10%) | 2 (20%) |
| Granuloma | | 1 (10%) | 1 (10%) | 1 (10%) |
| Hemorrhage | | | | 1 (10%) |
| Hepatodiaphragmatic nodule | 3 (30%) | | 1 (10%) | 1 (10%) |
| Inflammation, subacute | | 1 (10%) | 3 (30%) | 1 (10%) |
| Mixed cell focus | | 2 (20%) | 2 (20%) | |
| Bile duct, hyperplasia | 5 (50%) | 4 (40%) | 4 (40%) | |
| Hepatocyte, vacuolization cytoplasmic | 2 (20%) | | 1 (10%) | |
| Kupffer cell, pigmentation | 1 (10%) | | 1 (10%) | |
| Lobules, necrosis | | | | 2 (20%) |
| Mesentery | (4) | (2) | (5) | (1) |
| Accessory spleen | | | | 1 (100%) |
| Fibrosis | 1 (25%) | | 1 (20%) | |
| Hemorrhage | | | 1 (20%) | |
| Fat, necrosis | 4 (100%) | 2 (100%) | 2 (40%) | |
| Pancreas | (10) | (10) | (10) | (10) |
| Atrophy | 3 (30%) | 4 (40%) | 5 (50%) | 4 (40%) |
| Cytoplasmic alteration | | | 1 (10%) | 1 (10%) |
| **Cardiovascular System** | | | | |
| Heart | (10) | (10) | (10) | (10) |
| Cardiomyopathy | 8 (80%) | 4 (40%) | 6 (60%) | 6 (60%) |

[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 Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***15-Month Interim Evaluation*** (continued) | | | | |
| **Endocrine System** | | | | |
| Adrenal cortex | (10) | (10) | (10) | (10) |
| Accessory adrenal cortical nodule | 4 (40%) | 1 (10%) | 1 (10%) | 3 (30%) |
| Hyperplasia, focal | 1 (10%) | | 1 (10%) | |
| Hypertrophy, focal | | | | 1 (10%) |
| Pituitary gland | (9) | (10) | (10) | (10) |
| Pars distalis, angiectasis | | 1 (10%) | | |
| Pars distalis, cyst | 1 (11%) | | 1 (10%) | 1 (10%) |
| Pars distalis, hemorrhage | | | | 1 (10%) |
| Pars distalis, hyperplasia, focal | 3 (33%) | 1 (10%) | 2 (20%) | 2 (20%) |
| Thyroid gland | (10) | (10) | (10) | (10) |
| Ultimobranchial cyst | | | 1 (10%) | 2 (20%) |
| C-cell, hyperplasia | | | 1 (10%) | |
| Follicle, cyst | | | 1 (10%) | 1 (10%) |
| **Genital System** | | | | |
| Epididymis | (10) | (10) | (10) | (10) |
| Atypia cellular | 1 (10%) | 2 (20%) | 1 (10%) | 2 (20%) |
| Preputial gland | (10) | (10) | (10) | (10) |
| Inflammation, chronic | 6 (60%) | 6 (60%) | 5 (50%) | 6 (60%) |
| Inflammation, suppurative | 2 (20%) | | 1 (10%) | 1 (10%) |
| Prostate | (10) | (10) | (10) | (10) |
| Corpora amylacea | 1 (10%) | 2 (20%) | 5 (50%) | 7 (70%) |
| Inflammation, suppurative | 5 (50%) | 5 (50%) | 6 (60%) | 4 (40%) |
| Epithelium, hyperplasia | 1 (10%) | | 1 (10%) | |
| Testes | (10) | (10) | (10) | (10) |
| Interstitial cell, hyperplasia | 5 (50%) | 6 (60%) | 8 (80%) | 4 (40%) |
| **Hematopoietic System** | | | | |
| Bone marrow | (10) | (10) | (10) | (10) |
| Myelofibrosis | | 1 (10%) | | |
| Lymph node | (2) | | (2) | (1) |
| Deep cervical, hemorrhage | 1 (50%) | | | |
| Deep cervical, pigmentation | 1 (50%) | | | |
| Mediastinal, hemorrhage | 2 (100%) | | 2 (100%) | 1 (100%) |
| Mediastinal, pigmentation | 2 (100%) | | 2 (100%) | 1 (100%) |
| Lymph node, mandibular | (10) | (10) | (10) | (10) |
| Ectasia | | 1 (10%) | 1 (10%) | |
| Hemorrhage | 2 (20%) | | 1 (10%) | |
| Hyperplasia, lymphoid | 1 (10%) | 1 (10%) | | |
| Lymph node, mesenteric | (10) | (10) | (10) | (10) |
| Ectasia | 1 (10%) | | | |
| Hemorrhage | | | 2 (20%) | |
| Hyperplasia, lymphoid | | 1 (10%) | | |
| Spleen | (10) | (10) | (10) | (10) |
| Hematopoietic cell proliferation | 2 (20%) | | | 1 (10%) |
| Pigmentation, hemosiderin | 10 (100%) | 10 (100%) | 10 (100%) | 7 (70%) |
| Thymus | (10) | (10) | (10) | (9) |
| Cyst | 1 (10%) | | | |

TABLE A5
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| *15-Month Interim Evaluation* (continued) | | | | |
| **Integumentary System** | | | | |
| Skin | (10) | (10) | (10) | (10) |
| Inflammation, chronic | | | 1 (10%) | |
| **Musculoskeletal System** | | | | |
| Bone | (10) | (10) | (10) | (10) |
| Femur, osteopetrosis | 1 (10%) | | | |
| **Respiratory System** | | | | |
| Lung | (10) | (10) | (10) | (10) |
| Hemorrhage | 1 (10%) | | | |
| Infiltration cellular, histiocyte | 2 (20%) | | | 2 (20%) |
| Alveolar epithelium, hyperplasia | 1 (10%) | | 1 (10%) | 1 (10%) |
| Nose | (10) | (10) | (10) | (10) |
| Exudate | | 2 (20%) | 1 (10%) | |
| Foreign body | | 2 (20%) | 1 (10%) | |
| Fungus | | 1 (10%) | | |
| Mucosa, hyperplasia | | | 1 (10%) | |
| Mucosa, metaplasia, squamous | | 1 (10%) | | |
| **Special Senses System** | | | | |
| Eye | | | | (1) |
| Cataract | | | | 1 (100%) |
| Hemorrhage | | | | 1 (100%) |
| Retina, degeneration | | | | 1 (100%) |
| **Urinary System** | | | | |
| Kidney | (10) | (10) | (10) | (10) |
| Nephropathy | 10 (100%) | 10 (100%) | 10 (100%) | 9 (90%) |
| Renal tubule, pigmentation | 10 (100%) | 10 (100%) | 10 (100%) | 10 (100%) |
| *Systems Examined With No Lesions Observed* | | | | |
| **General Body System** | | | | |
| **Nervous System** | | | | |

TABLE A5
**Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**
(continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** | | | | |
| **Alimentary System** | | | | |
| Intestine large, colon | (44) | (48) | (49) | (48) |
| Dilatation | 1 (2%) | | | |
| Parasite metazoan | 5 (11%) | 3 (6%) | 4 (8%) | 4 (8%) |
| Intestine large, rectum | (49) | (50) | (50) | (50) |
| Parasite metazoan | 10 (20%) | 5 (10%) | 1 (2%) | 8 (16%) |
| Intestine large, cecum | (47) | (50) | (50) | (50) |
| Edema | 3 (6%) | 2 (4%) | 4 (8%) | |
| Parasite metazoan | | 1 (2%) | | 1 (2%) |
| Ulcer | 1 (2%) | | | 1 (2%) |
| Intestine small, duodenum | (50) | (50) | (50) | (50) |
| Erosion | | 1 (2%) | | |
| Liver | (50) | (50) | (50) | (50) |
| Angiectasis | | 2 (4%) | 5 (10%) | 2 (4%) |
| Basophilic focus | 22 (44%) | 20 (40%) | 18 (36%) | 14 (28%) |
| Clear cell focus | 15 (30%) | 8 (16%) | 7 (14%) | 8 (16%) |
| Cyst | | | | 2 (4%) |
| Degeneration, cystic | 7 (14%) | 7 (14%) | 10 (20%) | 6 (12%) |
| Eosinophilic focus | 3 (6%) | 8 (16%) | 7 (14%) | 4 (8%) |
| Granuloma | | | | 7 (14%) |
| Hematopoietic cell proliferation | | 2 (4%) | | |
| Hemorrhage | | | | 1 (2%) |
| Hepatodiaphragmatic nodule | 1 (2%) | 4 (8%) | | 7 (14%) |
| Inflammation, subacute | | | | 4 (8%) |
| Mixed cell focus | 6 (12%) | 3 (6%) | 6 (12%) | 7 (14%) |
| Bile duct, hyperplasia | 39 (78%) | 41 (82%) | 38 (76%) | 27 (54%) |
| Centrilobular, necrosis | 1 (2%) | 4 (8%) | | 2 (4%) |
| Hepatocyte, hyperplasia, focal | | 1 (2%) | | |
| Hepatocyte, vacuolization cytoplasmic | 6 (12%) | 2 (4%) | 2 (4%) | 4 (8%) |
| Kupffer cell, pigmentation | 1 (2%) | 1 (2%) | 5 (10%) | 6 (12%) |
| Lobules, necrosis | 2 (4%) | | 2 (4%) | 1 (2%) |
| Mesentery | (7) | (13) | (12) | (5) |
| Accessory spleen | | 3 (23%) | | 1 (20%) |
| Angiectasis | 1 (14%) | | | |
| Fat, necrosis | 5 (71%) | 8 (62%) | 9 (75%) | 3 (60%) |
| Pancreas | (50) | (49) | (50) | (50) |
| Atrophy | 29 (58%) | 33 (67%) | 27 (54%) | 29 (58%) |
| Edema | | | 3 (6%) | |
| Acinus, cytoplasmic alteration | 1 (2%) | | 1 (2%) | |
| Acinus, hyperplasia, focal | 4 (8%) | 7 (14%) | 9 (18%) | 12 (24%) |
| Salivary glands | (50) | (50) | (50) | (50) |
| Cytoplasmic alteration | | | 1 (2%) | |
| Stomach, forestomach | (50) | (50) | (50) | (50) |
| Edema | 1 (2%) | | 4 (8%) | 3 (6%) |
| Ulcer | 2 (4%) | 1 (2%) | 6 (12%) | 3 (6%) |
| Mucosa, hyperplasia | 2 (4%) | | 3 (6%) | 1 (2%) |
| Stomach, glandular | (50) | (50) | (50) | (50) |
| Edema | 2 (4%) | | 3 (6%) | |
| Erosion | 2 (4%) | 5 (10%) | 2 (4%) | 1 (2%) |
| Ulcer | 2 (4%) | | 1 (2%) | 1 (2%) |
| Tongue | | (1) | (1) | (1) |
| Epithelium, hyperplasia | | 1 (100%) | | 1 (100%) |

TABLE A5
**Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**
(continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Cardiovascular System** | | | | |
| Blood vessel | (50) | (50) | (50) | (50) |
| Hypertrophy | 2 (4%) | | 1 (2%) | |
| Inflammation, subacute | 2 (4%) | | 1 (2%) | |
| Thrombosis | | | 1 (2%) | |
| Heart | (50) | (50) | (50) | (50) |
| Cardiomyopathy | 32 (64%) | 32 (64%) | 24 (48%) | 34 (68%) |
| Thrombosis | 2 (4%) | 3 (6%) | 1 (2%) | 5 (10%) |
| Schwann cell, hyperplasia | | | 1 (2%) | |
| **Endocrine System** | | | | |
| Adrenal cortex | (50) | (50) | (50) | (50) |
| Accessory adrenal cortical nodule | 11 (22%) | 11 (22%) | 9 (18%) | 6 (12%) |
| Angiectasis | | 1 (2%) | 3 (6%) | 2 (4%) |
| Degeneration, fatty | 3 (6%) | 5 (10%) | 6 (12%) | 8 (16%) |
| Hemorrhage | 1 (2%) | | | 1 (2%) |
| Hyperplasia, focal | 7 (14%) | 6 (12%) | 10 (20%) | 4 (8%) |
| Hypertrophy | | 1 (2%) | | |
| Hypertrophy, focal | 2 (4%) | 2 (4%) | 5 (10%) | 1 (2%) |
| Vacuolization cytoplasmic | 1 (2%) | | | |
| Adrenal medulla | (50) | (50) | (50) | (50) |
| Hyperplasia | 6 (12%) | 12 (24%) | 17 (34%) | 12 (24%) |
| Islets, pancreatic | (50) | (49) | (50) | (50) |
| Hyperplasia | 3 (6%) | 2 (4%) | | 2 (4%) |
| Metaplasia, hepatocyte | | 1 (2%) | | |
| Parathyroid gland | (49) | (50) | (49) | (49) |
| Hyperplasia | | | 1 (2%) | 1 (2%) |
| Pituitary gland | (50) | (50) | (50) | (49) |
| Pars distalis, angiectasis | 3 (6%) | | 3 (6%) | |
| Pars distalis, cyst | 3 (6%) | 5 (10%) | 6 (12%) | 2 (4%) |
| Pars distalis, hyperplasia | 1 (2%) | 1 (2%) | 1 (2%) | 2 (4%) |
| Pars distalis, hyperplasia, focal | 7 (14%) | 11 (22%) | 9 (18%) | 5 (10%) |
| Pars distalis, necrosis | | | | 1 (2%) |
| Pars intermedia, angiectasis | 3 (6%) | | 2 (4%) | |
| Pars intermedia, cyst | 1 (2%) | 1 (2%) | 1 (2%) | |
| Pars intermedia, hyperplasia | | 1 (2%) | 1 (2%) | |
| Thyroid gland | (50) | (50) | (50) | (50) |
| Ultimobranchial cyst | 2 (4%) | 2 (4%) | 1 (2%) | 2 (4%) |
| C-cell, hyperplasia | 4 (8%) | 10 (20%) | 12 (24%) | 7 (14%) |
| Follicle, cyst | 2 (4%) | 2 (4%) | | 4 (8%) |
| **General Body System** | | | | |
| None | | | | |

TABLE A5
**Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**
(continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Genital System** | | | | |
| Epididymis | (50) | (50) | (50) | (50) |
| Atypia cellular | 28 (56%) | 26 (52%) | 29 (58%) | 37 (74%) |
| Hypospermia | 34 (68%) | 37 (74%) | 41 (82%) | 43 (86%) |
| Preputial gland | (50) | (50) | (50) | (50) |
| Ectasia | 11 (22%) | 14 (28%) | 20 (40%) | 14 (28%) |
| Hyperplasia | 1 (2%) | 2 (4%) | 2 (4%) | 1 (2%) |
| Inflammation, chronic | 25 (50%) | 17 (34%) | 20 (40%) | 17 (34%) |
| Inflammation, suppurative | 9 (18%) | 9 (18%) | 14 (28%) | 6 (12%) |
| Prostate | (50) | (50) | (50) | (50) |
| Corpora amylacea | 17 (34%) | 28 (56%) | 32 (64%) | 30 (60%) |
| Fibrosis | | | | 1 (2%) |
| Hemorrhage | | 1 (2%) | | |
| Inflammation, suppurative | 29 (58%) | 29 (58%) | 26 (52%) | 19 (38%) |
| Epithelium, hyperplasia | 4 (8%) | 6 (12%) | 5 (10%) | 3 (6%) |
| Seminal vesicle | (50) | (50) | (50) | (50) |
| Dilatation | 4 (8%) | 2 (4%) | | 1 (2%) |
| Testes | (50) | (50) | (50) | (50) |
| Mineralization | | 1 (2%) | | |
| Interstitial cell, hyperplasia | 4 (8%) | 7 (14%) | 4 (8%) | 6 (12%) |
| Seminiferous tubule, atrophy | 2 (4%) | 5 (10%) | 6 (12%) | 4 (8%) |
| **Hematopoietic System** | | | | |
| Bone marrow | (50) | (50) | (50) | (50) |
| Hypercellularity | 1 (2%) | 4 (8%) | 1 (2%) | |
| Myelofibrosis | 2 (4%) | 3 (6%) | 4 (8%) | 5 (10%) |
| Lymph node | (23) | (22) | (30) | (19) |
| Iliac, hemorrhage | 1 (4%) | | | |
| Iliac, hyperplasia, lymphoid | 1 (4%) | | | |
| Iliac, pigmentation | 1 (4%) | | | |
| Mediastinal, fibrosis | | | 1 (3%) | |
| Mediastinal, hemorrhage | 5 (22%) | 5 (23%) | | |
| Mediastinal, pigmentation | 5 (22%) | 7 (32%) | 10 (33%) | 5 (26%) |
| Pancreatic, ectasia | | | 1 (3%) | |
| Pancreatic, hemorrhage | | 1 (5%) | | 1 (5%) |
| Pancreatic, hyperplasia, lymphoid | | | 1 (3%) | |
| Pancreatic, pigmentation | | | 4 (13%) | 2 (11%) |
| Renal, hyperplasia, lymphoid | 1 (4%) | | | |
| Renal, pigmentation | 2 (9%) | 2 (9%) | 3 (10%) | 2 (11%) |
| Lymph node, mandibular | (49) | (49) | (50) | (50) |
| Congestion | 1 (2%) | 2 (4%) | | |
| Ectasia | 6 (12%) | 3 (6%) | 4 (8%) | 4 (8%) |
| Hemorrhage | 4 (8%) | 3 (6%) | 2 (4%) | 6 (12%) |
| Hyperplasia, lymphoid | 3 (6%) | 2 (4%) | 1 (2%) | 5 (10%) |
| Pigmentation | 2 (4%) | 5 (10%) | 1 (2%) | |
| Lymph node, mesenteric | (50) | (50) | (50) | (50) |
| Ectasia | 6 (12%) | 4 (8%) | 2 (4%) | 1 (2%) |
| Hemorrhage | 1 (2%) | 3 (6%) | | 4 (8%) |
| Hyperplasia, lymphoid | 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 Butyl Benzyl Phthalate (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| *2-Year Study* (continued) | | | | |
| **Hematopoietic System** (continued) | | | | |
| Spleen | (50) | (50) | (50) | (50) |
| Developmental malformation | | 1 (2%) | | |
| Fibrosis | 5 (10%) | 11 (22%) | 14 (28%) | 10 (20%) |
| Granuloma | | | | 1 (2%) |
| Hematopoietic cell proliferation | 2 (4%) | 8 (16%) | 5 (10%) | 7 (14%) |
| Hemorrhage | | 1 (2%) | | |
| Metaplasia, osseous | 1 (2%) | | | |
| Necrosis | 1 (2%) | | 1 (2%) | |
| Pigmentation, hemosiderin | 14 (28%) | 1 (2%) | 2 (4%) | 6 (12%) |
| Lymphoid follicle, atrophy | | 1 (2%) | | |
| Thymus | (49) | (48) | (50) | (48) |
| Cyst | | | 1 (2%) | |
| Hemorrhage | 1 (2%) | 1 (2%) | | |
| **Integumentary System** | | | | |
| Mammary gland | (45) | (49) | (47) | (48) |
| Hyperplasia | 10 (22%) | 16 (33%) | 10 (21%) | 12 (25%) |
| Skin | (50) | (50) | (50) | (50) |
| Acanthosis | | 2 (4%) | 2 (4%) | 10 (20%) |
| Cyst epithelial inclusion | | 1 (2%) | 1 (2%) | 1 (2%) |
| Edema | | | 1 (2%) | |
| Hemorrhage | | | | 1 (2%) |
| Hyperkeratosis | | 2 (4%) | 3 (6%) | 13 (26%) |
| Inflammation, chronic | | 1 (2%) | | 2 (4%) |
| Inflammation, chronic, focal | 1 (2%) | | | |
| Metaplasia, osseous | 1 (2%) | | | |
| Ulcer | | | | 3 (6%) |
| **Musculoskeletal System** | | | | |
| Bone | (50) | (50) | (50) | (50) |
| Cranium, hyperostosis | | | | 1 (2%) |
| Cranium, osteopetrosis | 1 (2%) | 1 (2%) | 1 (2%) | |
| Femur, osteopetrosis | | | 3 (6%) | 1 (2%) |
| Rib, osteopetrosis | | | 1 (2%) | |
| **Nervous System** | | | | |
| Brain | (50) | (50) | (50) | (50) |
| Compression | 4 (8%) | 5 (10%) | | 2 (4%) |
| Gliosis | | | | 1 (2%) |
| Hemorrhage | | | | 2 (4%) |
| Hydrocephalus | 2 (4%) | 1 (2%) | | 1 (2%) |
| Mineralization | 1 (2%) | | | 1 (2%) |
| Necrosis | | | 1 (2%) | 2 (4%) |
| Cerebellum, necrosis | | | | 1 (2%) |

TABLE A5
**Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Respiratory System** | | | | |
| Lung | (50) | (50) | (50) | (50) |
| Congestion | 1 (2%) | 2 (4%) | | 1 (2%) |
| Foreign body | | 1 (2%) | | |
| Hemorrhage | | 1 (2%) | | 1 (2%) |
| Infiltration cellular, histiocyte | 13 (26%) | 12 (24%) | 14 (28%) | 6 (12%) |
| Inflammation, subacute | 1 (2%) | 3 (6%) | 1 (2%) | |
| Inflammation, suppurative | 1 (2%) | | | |
| Metaplasia, osseous | 1 (2%) | 1 (2%) | | |
| Alveolar epithelium, hyperplasia | 7 (14%) | 2 (4%) | 6 (12%) | 2 (4%) |
| Nose | (50) | (50) | (50) | (50) |
| Exudate | 17 (34%) | 10 (20%) | 13 (26%) | 17 (34%) |
| Foreign body | 8 (16%) | 4 (8%) | 6 (12%) | 5 (10%) |
| Fungus | 12 (24%) | 5 (10%) | 8 (16%) | 11 (22%) |
| Mucosa, hyperplasia | 11 (22%) | 8 (16%) | 5 (10%) | 12 (24%) |
| Mucosa, metaplasia, squamous | 9 (18%) | 5 (10%) | 7 (14%) | 2 (4%) |
| **Special Senses System** | | | | |
| Eye | (1) | | | (1) |
| Atrophy | 1 (100%) | | | |
| **Urinary System** | | | | |
| Kidney | (50) | (50) | (50) | (50) |
| Cyst | | 2 (4%) | 3 (6%) | |
| Inflammation, suppurative | | 3 (6%) | 9 (18%) | 4 (8%) |
| Mineralization | | 1 (2%) | 2 (4%) | |
| Nephropathy | 48 (96%) | 47 (94%) | 50 (100%) | 48 (96%) |
| Papilla, inflammation, suppurative | | 1 (2%) | | |
| Papilla, necrosis | | 1 (2%) | | |
| Renal tubule, atrophy | | 1 (2%) | 1 (2%) | |
| Renal tubule, cytoplasmic alteration | | | | 1 (2%) |
| Renal tubule, dilatation | | 1 (2%) | | 1 (2%) |
| Renal tubule, hyperplasia | | | 2 (4%) | |
| Renal tubule, pigmentation | 49 (98%) | 48 (96%) | 50 (100%) | 50 (100%) |
| Transitional epithelium, hyperplasia | 6 (12%) | 10 (20%) | 6 (12%) | 1 (2%) |
| Urinary bladder | (50) | (50) | (50) | (50) |
| Hemorrhage | | 1 (2%) | | |
| Inflammation, suppurative | | 2 (4%) | | |
| Transitional epithelium, hyperplasia | | | | 2 (4%) |

# APPENDIX B
# SUMMARY OF LESIONS IN FEMALE RATS IN THE 2-YEAR FEED STUDY OF BUTYL BENZYL PHTHALATE


| | | |
|---|---|---|
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TABLE B1
**Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate[a]**

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Disposition Summary** | | | | |
| Animals initially in study | 60 | 60 | 60 | 60 |
| *15-Month interim evaluation* | 10 | 10 | 10 | 10 |
| Early deaths | | | | |
| Moribund | 23 | 17 | 16 | 20 |
| Natural deaths | 2 | 4 | 5 | 1 |
| Survivors | | | | |
| Terminal sacrifice | 25 | 29 | 29 | 29 |
| Animals examined microscopically | 60 | 60 | 60 | 60 |
| ***15-Month Interim Evaluation*** | | | | |
| **Alimentary System** | | | | |
| Liver | (10) | (10) | (10) | (10) |
| Hepatocellular adenoma | | | 1 (10%) | |
| **Endocrine System** | | | | |
| Pituitary gland | (10) | (10) | (10) | (10) |
| Pars distalis, adenoma | | 1 (10%) | 1 (10%) | |
| Thyroid gland | (10) | (10) | (10) | (10) |
| C-cell, adenoma | | 1 (10%) | | |
| C-cell, adenoma, multiple | 1 (10%) | | | |
| Follicular cell, adenoma | 1 (10%) | | | |
| **Genital System** | | | | |
| Uterus | (10) | (10) | (10) | (10) |
| Deciduoma benign | 1 (10%) | | | |
| Polyp stromal | 1 (10%) | 1 (10%) | | |
| **Integumentary System** | | | | |
| Mammary gland | (10) | (10) | (10) | (10) |
| Fibroadenoma | | 1 (10%) | 1 (10%) | |
| **Nervous System** | | | | |
| Brain | (10) | (10) | (10) | (10) |
| Astrocytoma malignant | | | 1 (10%) | |
| **Systemic Lesions** | | | | |
| Multiple organs[b] | (10) | (10) | (10) | (10) |
| Leukemia mononuclear | 1 (10%) | | | |
| ***Systems Examined With No Neoplasms Observed*** | | | | |
| **Cardiovascular System** | | | | |
| **General Body System** | | | | |
| **Hematopoietic System** | | | | |
| **Musculoskeletal 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 Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| *2-Year Study* | | | | |
| **Alimentary System** | | | | |
| Intestine large, cecum | (50) | (50) | (49) | (49) |
| Intestine small, jejunum | (50) | (49) | (50) | (49) |
| Intestine small, ileum | (49) | (50) | (50) | (50) |
| Liver | (50) | (50) | (50) | (50) |
| Hepatocellular adenoma | | | 1 (2%) | 2 (4%) |
| Mesentery | (10) | (8) | (8) | (6) |
| Leiomyosarcoma, metastatic, uterus | | | 1 (13%) | |
| Lipoma | | 1 (13%) | | |
| Liposarcoma | 1 (10%) | | | |
| Schwannoma malignant | | 1 (13%) | | |
| Oral mucosa | | (1) | | (1) |
| Squamous cell carcinoma | | 1 (100%) | | |
| Pancreas | (50) | (50) | (50) | (50) |
| Acinus, adenoma | | | | 2 (4%) |
| Salivary glands | (49) | (50) | (50) | (50) |
| Stomach, forestomach | (50) | (50) | (50) | (50) |
| Squamous cell papilloma | 1 (2%) | | | |
| Stomach, glandular | (50) | (50) | (50) | (50) |
| Tongue | (3) | | (1) | |
| Squamous cell papilloma | 2 (67%) | | | |
| **Cardiovascular System** | | | | |
| Heart | (50) | (50) | (50) | (50) |
| Schwannoma malignant | 1 (2%) | | | 1 (2%) |
| **Endocrine System** | | | | |
| Adrenal cortex | (50) | (50) | (50) | (50) |
| Adenoma | 1 (2%) | 5 (10%) | 1 (2%) | |
| Carcinoma | | | 1 (2%) | |
| Adrenal medulla | (49) | (49) | (50) | (50) |
| Ganglioneuroma | | 1 (2%) | | |
| Pheochromocytoma benign | 1 (2%) | 2 (4%) | | 1 (2%) |
| Islets, pancreatic | (50) | (50) | (50) | (50) |
| Adenoma | 1 (2%) | 1 (2%) | | |
| Carcinoma | | | 2 (4%) | |
| Pituitary gland | (49) | (50) | (48) | (50) |
| Pars distalis, adenoma | 19 (39%) | 26 (52%) | 25 (52%) | 13 (26%) |
| Pars distalis, adenoma, multiple | 1 (2%) | | | |
| Pars distalis, carcinoma | 2 (4%) | | | |
| Pars intermedia, adenoma | | | | 1 (2%) |
| Thyroid gland | (49) | (50) | (50) | (50) |
| C-cell, adenoma | 4 (8%) | 4 (8%) | 3 (6%) | 2 (4%) |
| C-cell, carcinoma | 1 (2%) | 2 (4%) | | |
| C-cell, carcinoma, multiple | | | | 1 (2%) |
| Follicular cell, carcinoma | | 2 (4%) | | |
| **General Body System** | | | | |
| Tissue NOS | (1) | | | (1) |

TABLE B1
**Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Genital System** | | | | |
| Clitoral gland | (50) | (50) | (50) | (50) |
| Adenoma | 3 (6%) | 4 (8%) | 4 (8%) | 4 (8%) |
| Carcinoma | 4 (8%) | 6 (12%) | 5 (10%) | |
| Ovary | (50) | (50) | (50) | (50) |
| Arrhenoblastoma NOS | | | 1 (2%) | |
| Granulosa cell tumor benign | 2 (4%) | | | |
| Uterus | (50) | (50) | (50) | (50) |
| Deciduoma benign | | | | 1 (2%) |
| Leiomyoma | 1 (2%) | | | |
| Leiomyosarcoma | 1 (2%) | | 1 (2%) | |
| Polyp stromal | 6 (12%) | 9 (18%) | 6 (12%) | 7 (14%) |
| Sarcoma stromal | | 1 (2%) | | |
| **Hematopoietic System** | | | | |
| Bone marrow | (50) | (50) | (50) | (50) |
| Lymph node | (12) | (11) | (16) | (15) |
| Lymph node, mandibular | (49) | (50) | (50) | (50) |
| Lymph node, mesenteric | (50) | (50) | (50) | (50) |
| Spleen | (50) | (50) | (50) | (50) |
| Thymus | (49) | (50) | (50) | (48) |
| Carcinoma, metastatic, thyroid gland | | | | 1 (2%) |
| Thymoma benign | | | 1 (2%) | |
| **Integumentary System** | | | | |
| Mammary gland | (49) | (50) | (50) | (50) |
| Adenoma | 2 (4%) | | | |
| Carcinoma | 2 (4%) | 1 (2%) | 2 (4%) | |
| Fibroadenoma | 28 (57%) | 30 (60%) | 31 (62%) | 11 (22%) |
| Skin | (50) | (50) | (50) | (50) |
| Basal cell adenoma | | | | 1 (2%) |
| Basal cell carcinoma | | | 1 (2%) | |
| Keratoacanthoma | 1 (2%) | 1 (2%) | | |
| Squamous cell papilloma | | | 1 (2%) | |
| Subcutaneous tissue, fibroma | 1 (2%) | 1 (2%) | 2 (4%) | |
| Subcutaneous tissue, lipoma | | | 1 (2%) | |
| **Musculoskeletal System** | | | | |
| None | | | | |
| **Nervous System** | | | | |
| Brain | (50) | (50) | (50) | (50) |
| Astrocytoma malignant | | | | 1 (2%) |
| Carcinoma | 1 (2%) | | | |
| Carcinoma, metastatic, pituitary gland | 2 (4%) | | | |

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Respiratory System** | | | | |
| Lung | (50) | (50) | (50) | (50) |
| Alveolar/bronchiolar adenoma | 2 (4%) | 1 (2%) | 1 (2%) | |
| Carcinoma, metastatic, mammary gland | | 1 (2%) | | |
| **Special Senses System** | | | | |
| None | | | | |
| **Urinary System** | | | | |
| Kidney | (50) | (50) | (50) | (50) |
| Hemangioma | | | | 1 (2%) |
| Renal tubule, carcinoma | 1 (2%) | | 1 (2%) | |
| Urinary bladder | (50) | (50) | (50) | (50) |
| Transitional epithelium, papilloma | 1 (2%) | | | 2 (4%) |
| **Systemic Lesions** | | | | |
| Multiple organs | (50) | (50) | (50) | (50) |
| Leukemia mononuclear | 21 (42%) | 20 (40%) | 21 (42%) | 19 (38%) |
| **Neoplasm Summary** | | | | |
| Total animals with primary neoplasms[c] | | | | |
| 15-Month interim evaluation | 4 | 4 | 3 | |
| 2-Year study | 49 | 48 | 45 | 42 |
| Total primary neoplasms | | | | |
| 15-Month interim evaluation | 5 | 4 | 4 | |
| 2-Year study | 112 | 120 | 112 | 70 |
| Total animals with benign neoplasms | | | | |
| 15-Month interim evaluation | 3 | 4 | 2 | |
| 2-Year study | 40 | 43 | 41 | 31 |
| Total benign neoplasms | | | | |
| 15-Month interim evaluation | 4 | 4 | 3 | |
| 2-Year study | 77 | 86 | 77 | 48 |
| Total animals with malignant neoplasms | | | | |
| 15-Month interim evaluation | 1 | | 1 | |
| 2-Year study | 30 | 31 | 27 | 22 |
| Total malignant neoplasms | | | | |
| 15-Month interim evaluation | 1 | | 1 | |
| 2-Year study | 35 | 34 | 34 | 22 |
| Total animals with metastatic neoplasms | | | | |
| 2-Year study | 2 | 1 | 1 | 1 |
| Total metastatic neoplasms | | | | |
| 2-Year study | 2 | 1 | 1 | 1 |
| Total animals with uncertain neoplasms - benign or malignant | | | | |
| 2-Year study | | | 1 | |
| Total uncertain neoplasms | | | | |
| 2-Year study | | | 1 | |

[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 Butyl Benzyl Phthalate: 0 ppm**

| Number of Days on Study | 0 3 5 5 5 5 6 6 6 6 6 7 7 7 7 7 7 7 7 7 7 7 7 7 |
|---|---|
| | 3 6 1 7 8 9 6 7 8 8 9 0 0 0 1 1 1 1 1 1 2 2 2 2 |
| | 9 8 2 0 7 8 8 0 1 4 1 2 2 3 1 3 6 7 7 9 5 5 5 6 |
| **Carcass ID Number** | 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 |
| | 0 0 1 4 4 0 2 3 4 3 4 1 1 2 0 2 0 0 1 2 4 0 1 2 3 |
| | 9 8 6 8 4 3 4 7 9 4 1 1 3 0 2 8 4 5 4 2 5 7 0 3 9 |
| **Alimentary System** | |
| Esophagus | + + + + M + + + + + + + + + + + + + + + + + + + |
| Intestine large, colon | + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine large, rectum | + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine large, cecum | + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine small, duodenum | + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine small, jejunum | + + + + + + + + + + + + + + + + + + + + + + + + |
| Intestine small, ileum | + + + + + + + + + + + + + + + + + + + + + + + + |
| Liver | + + + + + + + + + + + + + + + + + + + + + + + + |
| Mesentery | + + + + + + + |
| Liposarcoma | X |
| Pancreas | + + + + + + + + + + + + + + + + + + + + + + + + |
| Salivary glands | + + + + M + + + + + + + + + + + + + + + + + + + |
| Stomach, forestomach | + + + + + + + + + + + + + + + + + + + + + + + + |
| Squamous cell papilloma | X |
| Stomach, glandular | + + + + + + + + + + + + + + + + + + + + + + + + |
| Tongue | + |
| Squamous cell papilloma | |
| **Cardiovascular System** | |
| Blood vessel | + + + + + + + + + + + + + + + + + + + + + + + + |
| Heart | + + + + + + + + + + + + + + + + + + + + + + + + |
| Schwannoma malignant | |
| **Endocrine System** | |
| Adrenal cortex | + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenoma | X |
| Adrenal medulla | + + + + M + + + + + + + + + + + + + + + + + + + |
| Pheochromocytoma benign | X |
| Islets, pancreatic | + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenoma | |
| Parathyroid gland | + + + + M + + + + + + M + + + + + + + + + + + + |
| Pituitary gland | + + + + M + + + + + + + + + + + + + + + + + + + |
| Pars distalis, adenoma | X X X X X X X X X |
| Pars distalis, adenoma, multiple | |
| Pars distalis, carcinoma | X |
| Thyroid gland | + + + + M + + + + + + + + + + + + + + + + + + + |
| C-cell, adenoma | X X X |
| C-cell, carcinoma | |
| **General Body System** | |
| Tissue NOS | + |
| **Genital System** | |
| Clitoral gland | + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenoma | X |
| Carcinoma | X |
| Ovary | + + + + + + + + + + + + + + + + + + + + + + + + |
| Granulosa cell tumor benign | X |

+: 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 Butyl Benzyl Phthalate: 0 ppm (continued)

| Number of Days on Study | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 3 0 1 | 3 1 2 | 3 1 7 | 3 1 8 | 3 1 9 | 3 2 5 | 3 2 6 | 3 2 9 | 3 3 0 | 3 3 2 | 3 3 5 | 3 3 6 | 3 4 0 | 3 4 3 | 3 4 7 | 3 0 6 | 3 1 5 | 3 2 1 | 3 2 7 | 3 3 1 | 3 3 3 | 3 3 8 | 3 4 2 | 3 4 6 | 3 5 0 | 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 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | 49 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Mesentery | + | | + | | | + | | | + | | | + | | | | | | | | | | | | | | 10 |
| Liposarcoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Tongue | | | | | | | | | | | | + | + | | | | | | | | | | | | | 3 |
| Squamous cell papilloma | | | | | | | | | | | | X | X | | | | | | | | | | | | | 2 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Schwannoma malignant | | | | | | | | | | | | | | | | | | | | X | | | | | | 1 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pheochromocytoma benign | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | X | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 47 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pars distalis, adenoma | X | | X | | | | | X | X | X | | X | X | | | | | | X | | | X | | X | | 19 |
| Pars distalis, adenoma, multiple | | | | | | | | | | | | | | | X | | | | | | | | | | | 1 |
| Pars distalis, carcinoma | | | | | | | | | | | | | | | | | | | | X | | | | | | 2 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| C-cell, adenoma | | | | | | | | | | | | X | | | | | | | | | | | | | | 4 |
| C-cell, carcinoma | | | | | | | | | | | | | | | | | | | X | | | | | | | 1 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Tissue NOS | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | | | | | X | | | | | | | | | X | 3 |
| Carcinoma | | | | | X | | | | | | | X | | | | | | X | | | | | | | | 4 |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Granulosa cell tumor benign | | | | | | X | | | | | | | | | | | | | | | | | | | | 2 |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 0 ppm** (continued)

| Number of Days on Study | 0 3 5 5 5 5 6 6 6 6 6 7 7 7 7 7 7 7 7 7 7 7 7 7 7<br>3 6 1 7 8 9 6 7 8 8 9 0 0 0 1 1 1 1 1 1 1 2 2 2 2<br>9 8 2 0 7 8 8 0 1 4 1 2 2 3 1 3 6 7 7 7 9 5 5 5 6 |
|---|---|
| **Carcass ID Number** | 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3<br>0 0 1 4 4 0 2 3 4 3 4 1 1 2 0 2 0 0 1 2 4 0 1 2 3<br>9 8 6 8 4 3 4 7 9 4 1 1 3 0 2 8 4 5 4 2 5 7 0 3 9 |
| **Genital System** (continued) | |
| Uterus | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Leiomyoma | |
| Leiomyosarcoma | |
| Polyp stromal | X X X |
| **Hematopoietic System** | |
| Bone marrow | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Lymph node | + + + + + + + + + + + |
| Lymph node, mandibular | + + + + M + + + + + + + + + + + + + + + + + + + + |
| Lymph node, mesenteric | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Spleen | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Thymus | + + + + + + + + + + + + + + + + M + + + + + + + + |
| **Integumentary System** | |
| Mammary gland | M + + + + + + + + + + + + + + + + + + + + + + + + + |
| Adenoma | |
| Carcinoma | X |
| Fibroadenoma | X X X X X X X X X X X X X X X X |
| Skin | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Keratoacanthoma | |
| Subcutaneous tissue, fibroma | X |
| **Musculoskeletal System** | |
| Bone | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Skeletal muscle | + |
| **Nervous System** | |
| Brain | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Carcinoma | |
| Carcinoma, metastatic, pituitary gland | X |
| **Respiratory System** | |
| Lung | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Alveolar/bronchiolar adenoma | X |
| Nose | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Trachea | + + + + M + + + + + + + + + + + + + + + + + + + + |
| **Special Senses System** | |
| Eye | + + |
| **Urinary System** | |
| Kidney | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Renal tubule, carcinoma | X |
| Urinary bladder | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Transitional epithelium, papilloma | X |
| **Systemic Lesions** | |
| Multiple organs | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Leukemia mononuclear | X 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 Butyl Benzyl Phthalate: 0 ppm (continued)

| Number of Days on Study | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 3 0 1 | 3 1 2 | 3 1 7 | 3 1 8 | 3 1 9 | 3 2 5 | 3 2 6 | 3 2 9 | 3 3 0 | 3 3 2 | 3 3 5 | 3 3 6 | 3 4 0 | 3 4 3 | 3 4 7 | 3 0 6 | 3 1 5 | 3 1 2 | 3 2 7 | 3 1 1 | 3 3 3 | 3 3 8 | 3 4 2 | 3 4 6 | 3 5 0 | Total Tissues/ Tumors |
| **Genital System** (continued) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leiomyoma | | | | | | | | | | | | | | | | X | | | | | | | | | | 1 |
| Leiomyosarcoma | | | | | | | | | | | X | | | | | | | | | | | | | | | 1 |
| Polyp stromal | | | | | | | | | | | | X | | | X | | | | X | | | | | | | 6 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node | | | + | | | | | | | | | | | | | + | + | | | | | | | | | 12 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Adenoma | | | | | | | | X | | | | X | | | | | | | | | | | | | | 2 |
| Carcinoma | | | | X | | | | | | | | | | | | | | | | | | | | | | 2 |
| Fibroadenoma | | | X | X | | X | X | X | | | X | X | | | X | X | X | | X | | X | X | | | X | 28 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Keratoacanthoma | | | | | | | | | | X | | | | | | | | | | | | | | | | 1 |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Skeletal muscle | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | X | | | | 1 |
| Carcinoma, metastatic, pituitary gland | | | | | | | | | | | | | | | | | | | | | X | | | | | 2 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Alveolar/bronchiolar adenoma | | | | | | | | | | | X | | | | | | | | | | | | | | | 2 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Eye | | | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Renal tubule, carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Transitional epithelium, papilloma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leukemia mononuclear | | | X | | X | | | | | | | X | X | | | | X | | | X | | | X | | 21 |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm**

| Number of Days on Study | 2 4 7 | 4 0 5 | 4 2 4 | 5 2 9 | 5 9 3 | 6 0 6 | 6 0 6 | 6 1 1 | 6 1 8 | 6 6 3 | 6 8 2 | 6 8 9 | 6 8 8 | 7 0 1 | 7 0 3 | 7 1 8 | 7 2 2 | 7 2 5 | 7 2 5 | 7 2 5 | 7 2 5 | 7 2 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 4 0 1 | 3 9 0 | 4 1 5 | 3 6 2 | 3 6 8 | 3 7 9 | 3 8 2 | 4 0 2 | 3 6 1 | 3 8 8 | 3 9 2 | 3 7 2 | 3 9 4 | 3 9 7 | 3 9 9 | 3 8 9 | 3 6 4 | 3 6 3 | 3 6 7 | 3 7 6 | 4 0 0 | 3 9 9 | 3 6 5 | 3 6 6 | 3 7 0 | 3 7 3 |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, colon | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Mesentery | + | | | | | | + | | | | | | | | | + | | | | | | | | | |
| Lipoma | | | | | | | X | | | | | | | | | | | | | | | | | | | |
| Schwannoma malignant | X | | | | | | | | | | | | | | | | | | | | | | | | | |
| Oral mucosa | | | | | | | | | | | | | | | | + | | | | | | | | | |
| Squamous cell carcinoma | | | | | | | | | | | | | | | | X | | | | | | | | | |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | X | X | | | X | X | |
| Adrenal medulla | I | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Ganglioneuroma | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Pheochromocytoma benign | | | | | | | | | | | | | | | X | | X | | | | | | | | | |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pars distalis, adenoma | | | | X | X | | X | X | X | | X | X | X | X | X | X | | | | | X | | | | X | X |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | | | X | | | | | |
| C-cell, carcinoma | | | | | | | | | | | | X | | | | | | | | | | | | | | |
| Follicular cell, carcinoma | | | | | | | | | | | | | | X | | | | | | | | | | | | |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | X | | | X | | | | |
| Carcinoma | | | | | | | | | X | | | | | | | | | | | | | | | | | |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Polyp stromal | X | | | | | | | | | | | | | | | | | X | X | X | | | | | | |
| Sarcoma stromal | | | | | | | | | | | | | | | | | X | | | | | | | | | |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm**
(continued)

| Number of Days on Study | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 3 7 4 | 3 7 7 | 3 7 8 | 3 8 1 | 3 8 4 | 3 8 5 | 3 8 7 | 3 9 1 | 3 9 5 | 3 9 6 | 4 0 3 | 4 0 4 | 4 0 6 | 4 0 7 | 4 0 9 | 3 6 9 | 3 7 1 | 3 7 5 | 3 8 0 | 3 8 3 | 3 8 6 | 3 9 3 | 3 9 8 | 4 0 5 | 4 0 8 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Mesentery | | | + | | | | | + | + | | | | | | | | | | | + | | + | | | 8 |
| Lipoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Schwannoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Oral mucosa | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Squamous cell carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | X | | | | | | | | | | | | | | 5 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Ganglioneuroma | | | | | | | | | | | | | | | | | | | | | X | | | | | 1 |
| Pheochromocytoma benign | | | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | | | | | | | | | | X | | | | | 1 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pars distalis, adenoma | | | | X | | X | X | X | | | X | X | X | X | | | | | X | X | X | X | X | | X | 26 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | | | | | | | | | | X | | | | | | | X | | X | | | 4 |
| C-cell, carcinoma | | | | X | | | | | | | | | | | | | | | | | | | | | | 2 |
| Follicular cell, carcinoma | | X | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | X | | | | | | | | | | | | | | | X | | | | | | | 4 |
| Carcinoma | | X | | | | | X | | | | | X | | | | | | X | | | X | | | | | 6 |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Polyp stromal | | | | X | | X | X | | | | X | | | | | | | X | | | | | | | 9 |
| Sarcoma stromal | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |

**TABLE B2**
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 6,000 ppm**
(continued)

| Number of Days on Study | 2 4 4 5 5 6 6 6 6 6 6 6 7 7 7 7 7 7 7 7 7 7 7 7<br>4 0 2 2 9 0 0 1 1 6 8 8 0 0 1 2 2 2 2 2 3 3 3 3<br>7 5 4 9 3 6 6 1 8 3 2 9 1 3 8 2 5 5 5 5 6 6 6 6 6 |
|---|---|
| **Carcass ID Number** | 4 3 4 3 3 3 3 4 3 3 3 3 3 3 3 3 3 3 4 3 3 3 3 3<br>0 9 1 6 6 7 8 0 6 8 9 7 9 9 8 6 6 6 7 0 9 6 6 7 7<br>1 0 5 2 8 9 2 2 1 8 2 2 4 7 9 4 3 7 6 0 9 5 6 0 3 |
| **Hematopoietic System** | |
| Bone marrow | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Lymph node | + + + + + + + + + + + + + + + + + |
| Lymph node, mandibular | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Lymph node, mesenteric | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Spleen | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Thymus | + + + + + + + + + + + + + + + + + + + + + + + + + |
| **Integumentary System** | |
| Mammary gland | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Carcinoma | X |
| Fibroadenoma | X X X X X X X X X X X X X X X |
| Skin | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Keratoacanthoma | |
| Subcutaneous tissue, fibroma | X |
| **Musculoskeletal System** | |
| Bone | + + + + + + + + + + + + + + + + + + + + + + + + + |
| **Nervous System** | |
| Brain | + + + + + + + + + + + + + + + + + + + + + + + + + |
| **Respiratory System** | |
| Lung | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Alveolar/bronchiolar adenoma | X |
| Carcinoma, metastatic, mammary gland | X |
| Nose | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Trachea | + + + + + + + + + + + + + + + + + + + + + + + + + |
| **Special Senses System** | |
| None | |
| **Urinary System** | |
| Kidney | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Urinary bladder | + + + + + + + + + + + + + + + + + + + + + + + + + |
| **Systemic Lesions** | |
| Multiple organs | + + + + + + + + + + + + + + + + + + + + + + + + + |
| Leukemia mononuclear | X 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 Butyl Benzyl Phthalate: 6,000 ppm
(continued)

| Number of Days on Study | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 4 0 | 7 4 0 | 7 4 0 | 7 4 0 | 7 4 0 | 7 3 6 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 3 7 4 | 3 7 7 | 3 7 8 | 3 8 1 | 3 8 4 | 3 8 5 | 3 8 7 | 3 9 1 | 3 9 5 | 3 9 6 | 4 0 3 | 4 0 4 | 4 0 6 | 4 0 7 | 4 0 9 | 3 6 9 | 3 7 1 | 3 7 5 | 3 8 0 | 3 8 3 | 3 8 6 | 3 9 3 | 3 9 8 | 4 0 5 | 4 0 8 | Total Tissues/ Tumors | |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Lymph node | | | | | | | + | | | | | | | | + | + | | + | | | | | | | | 11 | |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 | |
| Fibroadenoma | X | | | X | X | | | X | X | | | X | | X | X | X | X | | X | X | X | X | X | X | | 30 | |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Keratoacanthoma | | | | | | | X | | | | | | | | | | | | | | | | | | | 1 | |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 | |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Alveolar/bronchiolar adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 | |
| Carcinoma, metastatic, mammary gland | | | | | | | | | | | | | | | | | | | | | | | | | | 1 | |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 | |
| Leukemia mononuclear | | | X | | | | | X | X | | | | | | X | X | | | | X | | | | X | X | 20 | |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm**

| Number of Days on Study | 2 8 5 | 4 7 9 | 5 0 9 | 5 4 4 | 5 4 4 | 5 5 1 | 5 7 2 | 6 0 7 | 6 4 6 | 6 4 7 | 6 4 7 | 6 6 6 | 6 7 0 | 7 0 3 | 7 2 2 | 7 2 4 | 7 2 5 | 7 2 5 | 7 2 6 | 7 2 6 | 7 3 3 | 7 3 6 | 7 3 6 | 7 3 6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 4 6 7 | 4 6 1 | 4 6 7 | 4 2 3 | 4 2 6 | 4 6 6 | 4 5 0 | 4 4 0 | 4 4 9 | 4 3 0 | 4 3 1 | 4 3 2 | 4 5 5 | 4 2 4 | 4 3 6 | 4 6 0 | 4 4 3 | 4 4 7 | 4 4 4 | 4 5 7 | 4 6 8 | 4 2 1 | 4 2 2 | 4 2 8 | 4 2 9 |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Hepatocellular adenoma | | | | | | | | | | | | | | | | | | | | | | | | |
| Mesentery | | | | | | | | + | + | | + | | + | | + | | | | + | | | + | | |
| Leiomyosarcoma, metastatic, uterus | | | | | | | | | X | | | | | | | | | | | | | | | |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Tongue | | | | | | | | | | | | | | | | | | | | | | | | |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | |
| Carcinoma | | | | | | | X | | | | | | | | | | | | | | | | | |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoma | | | | | | | | | | | | X | | | | | | | | | | | | |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | M | + | + | + | + | + | + | + |
| Pars distalis, adenoma | | | | | | | | X | | | | | X | | | | | X | X | X | X | | X | X |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | X | | | | | | | | | | | | | X | | |
| Carcinoma | | | | | | | X | | | | | | | | | X | | | | | | | X | |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Arrhenoblastoma NOS | | | | | | | | | | | | | | | | | | | | X | | | | |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Leiomyosarcoma | | | | | | | | | X | | | | | | | | | | | | | | | |
| Polyp stromal | | | | | | | | | | | | | | | X | X | | | X | | | X | | |

TABLE B2
Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm
(continued)

| Number of Days on Study | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 4 3 4 | 4 3 5 | 4 3 8 | 4 3 9 | 4 4 2 | 4 4 5 | 4 4 8 | 4 2 5 | 4 3 3 | 4 3 7 | 4 4 1 | 4 4 6 | 4 5 1 | 4 5 2 | 4 5 3 | 4 5 4 | 4 6 2 | 4 6 5 | 4 6 9 | 4 7 0 | 4 5 6 | 4 5 8 | 4 5 9 | 4 6 3 | 4 6 4 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, cecum | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Hepatocellular adenoma | | | | | | | | | | | | | | X | | | | | | | | | | | | 1 |
| Mesentery | | | + | | | | | | | | | | | | | | | | | | | | | | | 8 |
| Leiomyosarcoma, metastatic, uterus | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Tongue | | | | | | | | | | | | | | | | | | | | | | | + | | | 1 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | X | | | | | | | | | | | | | | | | | | 1 |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Carcinoma | X | | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Pars distalis, adenoma | X | X | X | | X | X | X | | | | | X | X | X | X | X | X | X | | | X | X | X | | X | 25 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | X | | | | | | X | | | | | | | | | | | | X | | | 3 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| None | | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | | | | | | | | X | X | | | | | | | | | | 4 |
| Carcinoma | | | | | | | | | | | X | | | | | | | | | | | | X | | | 5 |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Arrhenoblastoma NOS | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leiomyosarcoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Polyp stromal | | X | | | | | | | | | | | | | | X | | | | | | | | | | 6 |

**TABLE B2**
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 12,000 ppm**
(continued)

| Number of Days on Study | 2 8 5 | 4 7 9 | 5 0 9 | 5 4 4 | 5 4 4 | 5 5 1 | 5 7 2 | 6 0 7 | 6 4 6 | 6 4 7 | 6 4 7 | 6 6 6 | 6 7 0 | 7 0 3 | 7 2 2 | 7 2 4 | 7 2 5 | 7 2 5 | 7 2 6 | 7 2 6 | 7 3 3 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 4 6 7 | 4 6 1 | 4 2 7 | 4 2 3 | 4 2 6 | 4 6 6 | 4 5 0 | 4 4 0 | 4 4 9 | 4 3 0 | 4 3 1 | 4 3 2 | 4 5 5 | 4 2 4 | 4 3 6 | 4 6 0 | 4 4 3 | 4 4 7 | 4 4 4 | 4 5 7 | 4 6 8 | 4 2 1 | 4 2 2 | 4 2 8 | 4 2 9 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node | | + | + | | | | + | | | | + | + | + | | + | + | | | + | | + | | | | |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymoma benign | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Fibroadenoma | | | X | X | X | | | X | X | X | | | X | X | | X | X | X | X | X | X | X | X | |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Basal cell carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Squamous cell papilloma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, fibroma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Subcutaneous tissue, lipoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Alveolar/bronchiolar adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ear | | + | | | | | | | | | | | | | | | | | | | | | | | |
| Eye | | | | + | | | | | | | | | | | | | | | | | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Renal tubule, carcinoma | | | | | | | | | | | | | | | | X | | | | | | | | | |
| Ureter | | | | | | | | | + | | | | | | | | | | | | | | | | |
| 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 Butyl Benzyl Phthalate: 12,000 ppm
(continued)

| Number of Days on Study | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 6 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 4 3 4 | 4 3 5 | 4 3 8 | 4 3 9 | 4 4 2 | 4 4 5 | 4 4 8 | 4 2 5 | 4 3 3 | 4 3 7 | 4 4 1 | 4 4 6 | 4 5 1 | 4 5 2 | 4 5 3 | 4 5 4 | 4 6 2 | 4 6 5 | 4 6 9 | 4 7 0 | 4 5 6 | 4 5 8 | 4 5 9 | 4 6 3 | 4 6 4 | Total Tissues/ Tumors |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node | + | | | | | | + | | | | | | | | + | + | | | | | | | | + | + | 16 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymoma benign | | | | | | | | | | | | | | | | | | | | | X | | | | | 1 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Carcinoma | | | | | | | | | | | | X | | | X | | | | | | | | | | | 2 |
| Fibroadenoma | | X | X | | X | X | | X | X | | X | | | X | X | X | | | X | | X | | X | X | | 31 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Basal cell carcinoma | | | X | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Squamous cell papilloma | | | | | | | | | | | | | | | | X | | | | | | | | | | 1 |
| Subcutaneous tissue, fibroma | | | X | | | | | | | | | | | | X | | | | | | | | | | | 2 |
| Subcutaneous tissue, lipoma | | | | | | | | | | | | | | | | | | | | | X | | | | | 1 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Alveolar/bronchiolar adenoma | | X | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ear | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Eye | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Renal tubule, carcinoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Ureter | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leukemia mononuclear | X | | | | X | X | | X | | | X | | | X | X | | | | | | | | | X | X | 21 |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 24,000 ppm**

| Number of Days on Study | 4 0 4 | 4 5 2 | 4 5 2 | 4 5 2 | 4 8 7 | 4 8 7 | 5 5 1 | 5 9 2 | 6 0 7 | 6 0 7 | 6 1 1 | 6 8 1 | 6 8 4 | 6 8 4 | 6 8 4 | 7 0 3 | 7 0 3 | 7 0 3 | 7 1 7 | 7 2 2 | 7 2 5 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 4 9 6 | 4 8 6 | 4 9 4 | 5 0 2 | 4 8 5 | 5 0 6 | 4 8 7 | 5 1 6 | 4 9 3 | 5 2 0 | 4 8 1 | 5 1 2 | 4 0 5 | 5 2 6 | 5 2 8 | 5 0 4 | 5 2 2 | 5 2 5 | 5 1 0 | 4 8 9 | 4 9 0 | 4 8 3 | 4 9 1 | 4 9 7 | 5 0 0 |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine large, cecum | A | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, jejunum | A | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Hepatocellular adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mesentery | | + | | | | | + | | | | | | | | | | | | | | | | + | | + |
| Oral mucosa | | | | | | | | | | | | | | | | | | | | | | | | | |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Acinus, adenoma | | | | | | | | | | | | | | | | | | X | | | | | X | | |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Schwannoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pheochromocytoma benign | | | | | | | | | | | | | | | | | | | | | | | | | |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Parathyroid gland | + | + | M | + | + | + | + | + | + | + | + | + | + | + | M | + | + | + | + | + | + | + | + | + | + |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Pars distalis, adenoma | | | | | | | | X | | | | X | | X | | X | | | | X | X | | X | X | |
| Pars intermedia, adenoma | | | | | X | | | | | | | | | | | | | | | | | | | | |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | X |
| C-cell, carcinoma, multiple | | | X | | | | | | | | | | | | | | | | | | | | | | |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Tissue NOS | | | | | | | | | | | | | | | | | | + | | | | | | | |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Deciduoma benign | | | | | | | | | | | | | X | | | | | | | | | | | | |
| Polyp stromal | | | | | X | | | | | | | | | | | | | | | | | | | | |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 24,000 ppm**
(continued)

| Number of Days on Study | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 5 0 7 | 5 1 1 | 5 1 4 | 5 1 7 | 5 2 1 | 5 2 7 | 4 8 2 | 4 8 4 | 4 8 8 | 4 9 2 | 4 9 5 | 4 9 8 | 4 9 9 | 5 0 1 | 5 0 3 | 5 0 8 | 5 0 9 | 5 1 3 | 5 1 5 | 5 1 8 | 5 1 9 | 5 2 3 | 5 2 4 | 5 2 9 | 5 3 0 | Total Tissues/ Tumors |
| **Alimentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Esophagus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, colon | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, rectum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine large, cecum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Intestine small, duodenum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Intestine small, jejunum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 49 |
| Intestine small, ileum | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Liver | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Hepatocellular adenoma | | | | | | | | | | | | | | | X | | | | | | | X | | | | 2 |
| Mesentery | | | | | | | | | | + | | | | | | | | + | | | | | | | | 6 |
| Oral mucosa | | | | | | | | | | | | | | | | | | + | | | | | | | | 1 |
| Pancreas | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Acinus, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 2 |
| Salivary glands | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, forestomach | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Stomach, glandular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Cardiovascular System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Blood vessel | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Heart | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Schwannoma malignant | | | | | | | | | | | | | X | | | | | | | | | | | | | 1 |
| **Endocrine System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Adrenal cortex | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adrenal medulla | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pheochromocytoma benign | | | | | | | | | | | | | | | | X | | | | | | | | | | 1 |
| Islets, pancreatic | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Parathyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Pituitary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Pars distalis, adenoma | | | | | | | X | X | | | | | | | | | | X | | X | | | | X | | 13 |
| Pars intermedia, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Thyroid gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| C-cell, adenoma | | | | | | | | | | | | | | | | | | | | | | | | | X | 2 |
| C-cell, carcinoma, multiple | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **General Body System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Tissue NOS | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Genital System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Clitoral gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Adenoma | | | | | | | | X | X | | | | | | X | | | | | | | | | | X | 4 |
| Ovary | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Uterus | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Deciduoma benign | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Polyp stromal | | | X | | | | | | X | | | | | | | X | | | X | | | | X | X | 7 |

TABLE B2
**Individual Animal Tumor Pathology of Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate: 24,000 ppm**
(continued)

| Number of Days on Study | 4 0 4 | 4 5 2 | 4 5 2 | 4 5 2 | 4 8 7 | 4 8 7 | 5 5 1 | 5 9 2 | 6 0 7 | 6 0 7 | 6 1 1 | 6 8 1 | 6 8 4 | 6 8 4 | 6 8 4 | 7 0 3 | 7 0 3 | 7 0 3 | 7 1 7 | 7 2 2 | 7 2 5 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Carcass ID Number** | 4 9 6 | 4 8 6 | 4 9 4 | 5 0 2 | 4 8 5 | 5 0 6 | 4 8 7 | 5 1 6 | 4 9 3 | 5 2 0 | 4 8 1 | 5 1 2 | 5 0 5 | 5 2 6 | 5 2 8 | 5 0 4 | 2 2 | 5 2 5 | 5 1 0 | 4 8 9 | 4 9 0 | 4 8 3 | 4 9 1 | 4 9 7 | 5 0 0 |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node | + | + | | | | | + | | | + | + | | | + | + | | + | | + | | | | | | |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Thymus | + | I | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Carcinoma, metastatic, thyroid gland | | | X | | | | | | | | | | | | | | | | | | | | | | |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Fibroadenoma | | | | | X | X | | | | | X | X | X | | | | | X | | | | | | | |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Basal cell adenoma | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Skeletal muscle | | | | | | | | | | | | | | | | | | | | | | | | | |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Astrocytoma malignant | X | | | | | | | | | | | | | | | | | | | | | | | | |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Eye | | | | + | | | | | | | | | | | | | | | | + | | | | | |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Hemangioma | | | | | | | | | | | | | | | | | | | | | | | | | |
| Ureter | | | | | | | + | | | | | | | | | | | | | | | | | | |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Transitional epithelium, papilloma | | | | X | | | | | | | | | | | | | | | | | | | | | |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + |
| Leukemia mononuclear | | 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 Butyl Benzyl Phthalate: 24,000 ppm
(continued)

| Number of Days on Study | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 7 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | 7 3 8 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Carcass ID Number | 5 0 7 | 5 1 1 | 5 1 4 | 5 1 7 | 5 2 1 | 5 2 7 | 5 4 2 | 5 4 4 | 5 4 8 | 5 4 9 2 | 5 4 9 5 | 5 4 9 8 | 5 4 9 9 | 5 5 0 1 | 5 5 0 3 | 5 5 0 8 | 5 5 0 9 | 5 5 1 3 | 5 5 1 5 | 5 5 1 8 | 5 5 1 9 | 5 5 2 3 | 5 5 2 4 | 5 5 2 9 | 5 5 3 0 | Total Tissues/ Tumors |
| **Hematopoietic System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone marrow | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node | | | | | | | | + | | | + | + | | | | + | | | | + | | | | + | | 15 |
| Lymph node, mandibular | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Lymph node, mesenteric | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Spleen | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Thymus | + | M | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 48 |
| Carcinoma, metastatic, thyroid gland | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Integumentary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mammary gland | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Fibroadenoma | | | | | | | X | | | | X | | | X | | X | | | | | | | | | X | 11 |
| Skin | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Basal cell adenoma | | | | | | | | X | | | | | | | | | | | | | | | | | | 1 |
| **Musculoskeletal System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Bone | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Skeletal muscle | + | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Nervous System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Brain | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Astrocytoma malignant | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| **Respiratory System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Lung | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Nose | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Trachea | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| **Special Senses System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Eye | + | | | | | | | | | | | | | | | | | | | | | | | | | 3 |
| **Urinary System** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Kidney | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Hemangioma | | | | X | | | | | | | | | | | | | | | | | | | | | | 1 |
| Ureter | | | | | | | | | | | | | | | | | | | | | | | | | | 1 |
| Urinary bladder | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Transitional epithelium, papilloma | | | | | | | | | | | | | | | | | | | | | | | | | X | 2 |
| **Systemic Lesions** | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Multiple organs | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | + | 50 |
| Leukemia mononuclear | | | X | | | | | | X | | | X | | | | X | | X | | X | | | X | | X | 19 |

TABLE B3
**Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Adrenal Cortex: Adenoma** | | | | |
| Overall rate[a] | 1/50 (2%) | 5/50 (10%) | 1/50 (2%) | 0/50 (0%) |
| Adjusted rate[b] | 2.4% | 15.6% | 3.4% | 0.0% |
| Terminal rate[c] | 0/25 (0%) | 3/29 (10%) | 1/29 (3%) | 0/29 (0%) |
| First incidence (days) | 681 | 725 | 736 (T) | —[e] |
| Life table test[d] | P=0.123N | P=0.137 | P=0.758N | P=0.515N |
| Logistic regression test[d] | P=0.146N | P=0.101 | P=0.761 | P=0.499N |
| Cochran-Armitage test[d] | P=0.134N | | | |
| Fisher exact test[d] | | P=0.102 | P=0.753N | P=0.500N |
| **Clitoral Gland: Adenoma** | | | | |
| Overall rate | 3/50 (6%) | 4/50 (8%) | 4/50 (8%) | 4/50 (8%) |
| Adjusted rate | 11.2% | 13.0% | 12.5% | 13.8% |
| Terminal rate | 2/25 (8%) | 3/29 (10%) | 3/29 (10%) | 4/29 (14%) |
| First incidence (days) | 725 | 725 | 646 | 736 (T) |
| Life table test | P=0.495 | P=0.582 | P=0.566 | P=0.572 |
| Logistic regression test | P=0.409 | P=0.541 | P=0.489 | P=0.530 |
| Cochran-Armitage test | P=0.446 | | | |
| Fisher exact test | | P=0.500 | P=0.500 | P=0.500 |
| **Clitoral Gland: Carcinoma** | | | | |
| Overall rate | 4/50 (8%) | 6/50 (12%) | 5/50 (10%) | 0/50 (0%) |
| Adjusted rate | 14.3% | 19.2% | 14.9% | 0.0% |
| Terminal rate | 3/25 (12%) | 5/29 (17%) | 3/29 (10%) | 0/29 (0%) |
| First incidence (days) | 702 | 618 | 572 | — |
| Life table test | P=0.037N | P=0.439 | P=0.567 | P=0.054N |
| Logistic regression test | P=0.052N | P=0.363 | P=0.492 | P=0.069N |
| Cochran-Armitage test | P=0.047N | | | |
| Fisher exact test | | P=0.370 | P=0.500 | P=0.059N |
| **Clitoral Gland: Adenoma or Carcinoma** | | | | |
| Overall rate | 7/50 (14%) | 10/50 (20%) | 9/50 (18%) | 4/50 (8%) |
| Adjusted rate | 24.7% | 31.4% | 26.5% | 13.8% |
| Terminal rate | 5/25 (20%) | 8/29 (28%) | 6/29 (21%) | 4/29 (14%) |
| First incidence (days) | 702 | 618 | 572 | 736 (T) |
| Life table test | P=0.119N | P=0.402 | P=0.491 | P=0.198N |
| Logistic regression test | P=0.188N | P=0.291 | P=0.377 | P=0.265N |
| Cochran-Armitage test | P=0.158N | | | |
| Fisher exact test | | P=0.298 | P=0.393 | P=0.262N |
| **Mammary Gland: Fibroadenoma** | | | | |
| Overall rate | 28/50 (56%) | 30/50 (60%) | 31/50 (62%) | 11/50 (22%) |
| Adjusted rate | 71.0% | 72.8% | 71.7% | 28.9% |
| Terminal rate | 14/25 (56%) | 18/29 (62%) | 17/29 (59%) | 5/29 (17%) |
| First incidence (days) | 587 | 606 | 509 | 487 |
| Life table test | P<0.001N | P=0.494N | P=0.557 | P=0.001N |
| Logistic regression test | P<0.001N | P=0.379 | P=0.318 | P<0.001N |
| Cochran-Armitage test | P<0.001N | | | |
| Fisher exact test | | P=0.420 | P=0.342 | P<0.001N |

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Mammary Gland: Fibroadenoma or Adenoma** | | | | |
| Overall rate | 28/50 (56%) | 30/50 (60%) | 31/50 (62%) | 11/50 (22%) |
| Adjusted rate | 71.0% | 72.8% | 71.7% | 28.9% |
| Terminal rate | 14/25 (56%) | 18/29 (62%) | 17/29 (59%) | 5/29 (17%) |
| First incidence (days) | 587 | 606 | 509 | 487 |
| Life table test | P<0.001N | P=0.494N | P=0.557 | P=0.001N |
| Logistic regression test | P<0.001N | P=0.379 | P=0.318 | P<0.001N |
| Cochran-Armitage test | P<0.001N | | | |
| Fisher exact test | | P=0.420 | P=0.342 | P<0.001N |
| **Mammary Gland: Adenoma or Carcinoma** | | | | |
| Overall rate | 4/50 (8%) | 1/50 (2%) | 2/50 (4%) | 0/50 (0%) |
| Adjusted rate | 14.6% | 2.1% | 6.9% | 0.0% |
| Terminal rate | 3/25 (12%) | 0/29 (0%) | 2/29 (7%) | 0/29 (0%) |
| First incidence (days) | 716 | 424 | 736 (T) | — |
| Life table test | P=0.044N | P=0.154N | P=0.277N | P=0.052N |
| Logistic regression test | P=0.052N | P=0.180N | P=0.304N | P=0.064N |
| Cochran-Armitage test | P=0.052N | | | |
| Fisher exact test | | P=0.181N | P=0.339N | P=0.059N |
| **Mammary Gland: Fibroadenoma, Adenoma, or Carcinoma** | | | | |
| Overall rate | 29/50 (58%) | 31/50 (62%) | 32/50 (64%) | 11/50 (22%) |
| Adjusted rate | 71.8% | 73.3% | 74.0% | 28.9% |
| Terminal rate | 14/25 (56%) | 18/29 (62%) | 18/29 (62%) | 5/29 (17%) |
| First incidence (days) | 587 | 424 | 509 | 487 |
| Life table test | P<0.001N | P=0.492N | P=0.560N | P<0.001N |
| Logistic regression test | P<0.001N | P=0.392 | P=0.313 | P<0.001N |
| Cochran-Armitage test | P<0.001N | | | |
| Fisher exact test | | P=0.419 | P=0.341 | P<0.001N |
| **Pituitary Gland (Pars Distalis): Adenoma** | | | | |
| Overall rate | 20/49 (41%) | 26/50 (52%) | 25/48 (52%) | 13/50 (26%) |
| Adjusted rate | 57.0% | 63.4% | 71.2% | 36.1% |
| Terminal rate | 11/25 (44%) | 15/29 (52%) | 19/29 (66%) | 7/29 (24%) |
| First incidence (days) | 598 | 529 | 607 | 592 |
| Life table test | P=0.035N | P=0.316 | P=0.396 | P=0.086N |
| Logistic regression test | P=0.053N | P=0.168 | P=0.127 | P=0.121N |
| Cochran-Armitage test | P=0.036N | | | |
| Fisher exact test | | P=0.180 | P=0.182 | P=0.088N |
| **Pituitary Gland (Pars Distalis): Adenoma or Carcinoma** | | | | |
| Overall rate | 22/49 (45%) | 26/50 (52%) | 25/48 (52%) | 13/50 (26%) |
| Adjusted rate | 61.3% | 63.4% | 71.2% | 36.1% |
| Terminal rate | 12/25 (48%) | 15/29 (52%) | 19/29 (66%) | 7/29 (24%) |
| First incidence (days) | 598 | 529 | 607 | 592 |
| Life table test | P=0.018N | P=0.455 | P=0.553 | P=0.042N |
| Logistic regression test | P=0.025N | P=0.290 | P=0.232 | P=0.058N |
| Cochran-Armitage test | P=0.016N | | | |
| Fisher exact test | | P=0.307 | P=0.307 | P=0.039N |

TABLE B3
**Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Thyroid Gland (C-cell): Adenoma** | | | | |
| Overall rate | 4/49 (8%) | 4/50 (8%) | 3/50 (6%) | 2/50 (4%) |
| Adjusted rate | 11.4% | 13.0% | 10.3% | 6.9% |
| Terminal rate | 1/25 (4%) | 3/29 (10%) | 3/29 (10%) | 2/29 (7%) |
| First incidence (days) | 702 | 725 | 736 (T) | 736 (T) |
| Life table test | P=0.209N | P=0.596N | P=0.463N | P=0.326N |
| Logistic regression test | P=0.256N | P=0.644 | P=0.507N | P=0.359N |
| Cochran-Armitage test | P=0.227N | | | |
| Fisher exact test | | P=0.631N | P=0.489N | P=0.329N |
| **Thyroid Gland (C-cell): Adenoma or Carcinoma** | | | | |
| Overall rate | 5/49 (10%) | 6/50 (12%) | 3/50 (6%) | 3/50 (6%) |
| Adjusted rate | 15.1% | 18.5% | 10.3% | 8.8% |
| Terminal rate | 2/25 (8%) | 4/29 (14%) | 3/29 (10%) | 2/29 (7%) |
| First incidence (days) | 702 | 689 | 736 (T) | 452 |
| Life table test | P=0.186N | P=0.557 | P=0.316N | P=0.337N |
| Logistic regression test | P=0.217N | P=0.499 | P=0.363N | P=0.346N |
| Cochran-Armitage test | P=0.200N | | | |
| Fisher exact test | | P=0.514 | P=0.346N | P=0.346N |
| **Uterus: Stromal Polyp** | | | | |
| Overall rate | 6/50 (12%) | 9/50 (18%) | 6/50 (12%) | 7/50 (14%) |
| Adjusted rate | 19.6% | 26.1% | 18.0% | 22.4% |
| Terminal rate | 3/25 (12%) | 5/29 (17%) | 3/29 (10%) | 6/29 (21%) |
| First incidence (days) | 681 | 247 | 722 | 487 |
| Life table test | P=0.498N | P=0.387 | P=0.523N | P=0.571 |
| Logistic regression test | P=0.535 | P=0.287 | P=0.611 | P=0.470 |
| Cochran-Armitage test | P=0.555 | | | |
| Fisher exact test | | P=0.288 | P=0.620N | P=0.500 |
| **Uterus: Stromal Polyp or Stromal Sarcoma** | | | | |
| Overall rate | 6/50 (12%) | 10/50 (20%) | 6/50 (12%) | 7/50 (14%) |
| Adjusted rate | 19.6% | 28.2% | 18.0% | 22.4% |
| Terminal rate | 3/25 (12%) | 5/29 (17%) | 3/29 (10%) | 6/29 (21%) |
| First incidence (days) | 681 | 247 | 722 | 487 |
| Life table test | P=0.458N | P=0.304 | P=0.523N | P=0.571 |
| Logistic regression test | P=0.535N | P=0.206 | P=0.611 | P=0.470 |
| Cochran-Armitage test | P=0.514N | | | |
| Fisher exact test | | P=0.207 | P=0.620N | P=0.500 |
| **All Organs: Mononuclear Cell Leukemia** | | | | |
| Overall rate | 21/50 (42%) | 20/50 (40%) | 21/50 (42%) | 19/50 (38%) |
| Adjusted rate | 51.7% | 46.6% | 52.0% | 46.1% |
| Terminal rate | 7/25 (28%) | 8/29 (28%) | 10/29 (34%) | 8/29 (28%) |
| First incidence (days) | 368 | 405 | 509 | 452 |
| Life table test | P=0.382N | P=0.402N | P=0.436N | P=0.398N |
| Logistic regression test | P=0.340N | P=0.500N | P=0.578 | P=0.422N |
| Cochran-Armitage test | P=0.394N | | | |
| Fisher exact test | | P=0.500N | P=0.580N | P=0.419N |

TABLE B3
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **All Organs: Benign Neoplasms** | | | | |
| Overall rate | 40/50 (80%) | 43/50 (86%) | 41/50 (82%) | 31/50 (62%) |
| Adjusted rate | 88.7% | 91.4% | 93.1% | 74.9% |
| Terminal rate | 20/25 (80%) | 25/29 (86%) | 26/29 (90%) | 19/29 (66%) |
| First incidence (days) | 512 | 247 | 509 | 452 |
| Life table test | P=0.028N | P=0.500N | P=0.377N | P=0.060N |
| Logistic regression test | P=0.009N | P=0.287 | P=0.457 | P=0.047N |
| Cochran-Armitage test | P=0.007N | | | |
| Fisher exact test | | P=0.298 | P=0.500 | P=0.038N |
| **All Organs: Malignant Neoplasms** | | | | |
| Overall rate | 30/50 (60%) | 31/50 (62%) | 27/50 (54%) | 22/50 (44%) |
| Adjusted rate | 70.1% | 66.3% | 63.8% | 50.7% |
| Terminal rate | 13/25 (52%) | 14/29 (48%) | 14/29 (48%) | 9/29 (31%) |
| First incidence (days) | 39 | 247 | 509 | 404 |
| Life table test | P=0.068N | P=0.466N | P=0.240N | P=0.110N |
| Logistic regression test | P=0.032N | P=0.516 | P=0.343N | P=0.067N |
| Cochran-Armitage test | P=0.038N | | | |
| Fisher exact test | | P=0.500 | P=0.343N | P=0.080N |
| **All Organs: Benign or Malignant Neoplasms** | | | | |
| Overall rate | 49/50 (98%) | 48/50 (96%) | 45/50 (90%) | 42/50 (84%) |
| Adjusted rate | 98.0% | 96.0% | 93.8% | 84.0% |
| Terminal rate | 24/25 (96%) | 27/29 (93%) | 26/29 (90%) | 21/29 (72%) |
| First incidence (days) | 39 | 247 | 509 | 404 |
| Life table test | P=0.092N | P=0.252N | P=0.145N | P=0.123N |
| Logistic regression test | P=0.004N | P=0.552N | P=0.087N | P=0.027N |
| Cochran-Armitage test | P=0.004N | | | |
| Fisher exact test | | P=0.500N | P=0.102N | P=0.015N |

(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, 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 Pancreatic Neoplasms in Untreated Female F344/N Rats[a]**

| | Incidence in Controls | | |
|---|---|---|---|
| | Adenoma | Carcinoma | Adenoma or Carcinoma |
| **Overall Historical Incidence** | | | |
| Total | 2/1,194 (0.2%) | 1/1,194 (0.1%) | 3/1,194 (0.3%) |
| Standard deviation | 0.8% | 0.4% | 0.9% |
| Range | 0%-4% | 0%-2% | 0%-4% |

[a] Data as of 17 June 1994

TABLE B4b
**Historical Incidence of Urinary Bladder Papilloma in Untreated Female F344/N Rats[a]**

| | Incidence in Controls |
|---|---|
| **Overall Historical Incidence** | |
| Total | 4/1,182 (0.3%) |
| Standard deviation | 0.8% |
| Range | 0%-2% |

[a] Data as of 17 June 1994

TABLE B4c
**Historical Incidence of Mammary Gland Fibroadenoma in Untreated Female F344/N Rats[a]**

| | Incidence in Controls |
|---|---|
| **Overall Historical Incidence** | |
| Total | 465/1,202 (38.7%) |
| Standard deviation | 12.7% |
| Range | 8%-58% |

[a] Data as of 17 June 1994

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Disposition Summary** | | | | |
| Animals initially in study | 60 | 60 | 60 | 60 |
| *15-Month interim evaluation* | 10 | 10 | 10 | 10 |
| Early deaths | | | | |
| Moribund | 23 | 17 | 16 | 20 |
| Natural deaths | 2 | 4 | 5 | 1 |
| Survivors | | | | |
| Terminal sacrifice | 25 | 29 | 29 | 29 |
| Animals examined microscopically | 60 | 60 | 60 | 60 |
| ***15-Month Interim Evaluation*** | | | | |
| **Alimentary System** | | | | |
| Intestine large, colon | (10) | (10) | (10) | (10) |
| Parasite metazoan | | | | 1 (10%) |
| Intestine large, cecum | (9) | (10) | (10) | (10) |
| Parasite metazoan | | 1 (10%) | | |
| Liver | (10) | (10) | (10) | (10) |
| Basophilic focus | 10 (100%) | 10 (100%) | 8 (80%) | 9 (90%) |
| Eosinophilic focus | | 1 (10%) | 1 (10%) | |
| Granuloma | 1 (10%) | 3 (30%) | 3 (30%) | |
| Hepatodiaphragmatic nodule | 1 (10%) | 1 (10%) | 2 (20%) | |
| Bile duct, hyperplasia | 1 (10%) | | | |
| Kupffer cell, pigmentation | | | 1 (10%) | |
| Mesentery | (1) | | (4) | |
| Accessory spleen | | | 1 (25%) | |
| Fat, necrosis | 1 (100%) | | 3 (75%) | |
| Pancreas | (10) | (10) | (10) | (10) |
| Atrophy | 1 (10%) | 2 (20%) | | |
| Tongue | | | | (1) |
| Epithelium, hyperplasia | | | | 1 (100%) |
| **Cardiovascular System** | | | | |
| Heart | (10) | (10) | (10) | (10) |
| Cardiomyopathy | 1 (10%) | 1 (10%) | 1 (10%) | |
| **Endocrine System** | | | | |
| Adrenal cortex | (10) | (10) | (10) | (10) |
| Accessory adrenal cortical nodule | 2 (20%) | 1 (10%) | 2 (20%) | 4 (40%) |
| Angiectasis | 5 (50%) | 6 (60%) | 7 (70%) | 3 (30%) |
| Cyst | | | | 1 (10%) |
| Degeneration, fatty | 1 (10%) | 1 (10%) | | |
| Hyperplasia, focal | | | | 1 (10%) |
| Hypertrophy, focal | 1 (10%) | 1 (10%) | 2 (20%) | |
| Parathyroid gland | (9) | (10) | (10) | (10) |
| Cyst | | | 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 Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| ***15-Month Interim Evaluation*** (continued) | | | | |
| **Endocrine System** (continued) | | | | |
| Pituitary gland | (10) | (10) | (10) | (10) |
| Pars distalis, angiectasis | 1 (10%) | 2 (20%) | 1 (10%) | |
| Pars distalis, cyst | 6 (60%) | 10 (100%) | 8 (80%) | 1 (10%) |
| Pars distalis, hemorrhage | | | 1 (10%) | |
| Pars distalis, hyperplasia, focal | 1 (10%) | 2 (20%) | | 1 (10%) |
| Pars intermedia, cyst | | 1 (10%) | | |
| Thyroid gland | (10) | (10) | (10) | (10) |
| Ultimobranchial cyst | 1 (10%) | 3 (30%) | | |
| C-cell, hyperplasia | | 1 (10%) | 1 (10%) | |
| **Genital System** | | | | |
| Clitoral gland | (10) | (10) | (10) | (10) |
| Ectasia | | | | 1 (10%) |
| Inflammation, chronic | 2 (20%) | 2 (20%) | 1 (10%) | |
| Ovary | (10) | (10) | (10) | (10) |
| Cyst | | | | 1 (10%) |
| Uterus | (10) | (10) | (10) | (10) |
| Hydrometra | 2 (20%) | 3 (30%) | 2 (20%) | 2 (20%) |
| Hyperplasia, cystic | | | | 1 (10%) |
| Inflammation, suppurative | 1 (10%) | | | |
| **Hematopoietic System** | | | | |
| Lymph node | | (1) | (1) | (1) |
| Mediastinal, hemorrhage | | 1 (100%) | 1 (100%) | 1 (100%) |
| Mediastinal, pigmentation | | 1 (100%) | 1 (100%) | 1 (100%) |
| Lymph node, mandibular | (10) | (10) | (10) | (10) |
| Ectasia | 1 (10%) | 1 (10%) | 2 (20%) | 2 (20%) |
| Hemorrhage | | | | 1 (10%) |
| Hyperplasia, lymphoid | 2 (20%) | 1 (10%) | 1 (10%) | |
| Pigmentation | 3 (30%) | 4 (40%) | 4 (40%) | 4 (40%) |
| Spleen | (10) | (10) | (10) | (10) |
| Hematopoietic cell proliferation | 1 (10%) | 3 (30%) | 3 (30%) | 3 (30%) |
| Pigmentation, hemosiderin | 10 (100%) | 10 (100%) | 10 (100%) | 10 (100%) |
| Thymus | (9) | (9) | (10) | (10) |
| Hemorrhage | | 1 (11%) | | |
| **Integumentary System** | | | | |
| Mammary gland | (10) | (10) | (10) | (10) |
| Hyperplasia | 3 (30%) | 2 (20%) | 3 (30%) | 1 (10%) |
| **Musculoskeletal System** | | | | |
| Bone | (10) | (10) | (10) | (10) |
| Cranium, hyperostosis | | 1 (10%) | | |
| Cranium, osteopetrosis | | 1 (10%) | 1 (10%) | 1 (10%) |
| Femur, osteopetrosis | | 1 (10%) | 1 (10%) | 2 (20%) |

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| ***15-Month Interim Evaluation*** (continued) | | | | |
| **Respiratory System** | | | | |
| Lung | (10) | (10) | (10) | (10) |
| Infiltration cellular, histiocyte | 2 (20%) | 1 (10%) | 3 (30%) | 2 (20%) |
| Nose | (10) | (10) | (10) | (10) |
| Exudate | 1 (10%) | 1 (10%) | | |
| Foreign body | | 2 (20%) | | |
| Fungus | 1 (10%) | | | |
| Mucosa, metaplasia, squamous | 1 (10%) | | | |
| **Urinary System** | | | | |
| Kidney | (10) | (10) | (10) | (10) |
| Cyst | 1 (10%) | 2 (20%) | | |
| Mineralization | 10 (100%) | 9 (90%) | 9 (90%) | 8 (80%) |
| Nephropathy | 7 (70%) | 10 (100%) | 10 (100%) | 10 (100%) |
| Renal tubule, atrophy | 1 (10%) | | | |
| Renal tubule, pigmentation | 10 (100%) | 10 (100%) | 10 (100%) | 10 (100%) |
| ***Systems Examined With No Lesions Observed*** | | | | |
| **General Body System** | | | | |
| **Nervous System** | | | | |
| **Special Senses System** | | | | |
| ***2-Year Study*** | | | | |
| **Alimentary System** | | | | |
| Intestine large, colon | (50) | (48) | (49) | (50) |
| Parasite metazoan | 3 (6%) | 2 (4%) | 3 (6%) | 3 (6%) |
| Intestine large, rectum | (50) | (50) | (50) | (50) |
| Parasite metazoan | 3 (6%) | 5 (10%) | 11 (22%) | 4 (8%) |
| Intestine large, cecum | (50) | (50) | (49) | (49) |
| Edema | 2 (4%) | | 2 (4%) | 1 (2%) |
| Parasite metazoan | 2 (4%) | | 1 (2%) | 1 (2%) |
| Ulcer | | | 1 (2%) | |
| Intestine small, ileum | (49) | (50) | (50) | (50) |
| Inflammation, chronic | | 1 (2%) | | |
| Ulcer | 1 (2%) | | | |
| Liver | (50) | (50) | (50) | (50) |
| Angiectasis | | 1 (2%) | 4 (8%) | |
| Basophilic focus | 37 (74%) | 38 (76%) | 41 (82%) | 39 (78%) |
| Clear cell focus | 3 (6%) | 3 (6%) | 8 (16%) | 9 (18%) |
| Cyst | | 1 (2%) | 2 (4%) | 2 (4%) |
| Developmental malformation | 1 (2%) | | | |
| Eosinophilic focus | 19 (38%) | 15 (30%) | 9 (18%) | 20 (40%) |
| Granuloma | 6 (12%) | 11 (22%) | 6 (12%) | 2 (4%) |
| Hematopoietic cell proliferation | 1 (2%) | | | 1 (2%) |
| Hepatodiaphragmatic nodule | 9 (18%) | 6 (12%) | 6 (12%) | 12 (24%) |
| Inflammation, subacute | 3 (6%) | | | |
| Mixed cell focus | 10 (20%) | 4 (8%) | 5 (10%) | 6 (12%) |
| Thrombosis | | | 1 (2%) | |
| Bile duct, hyperplasia | 10 (20%) | 12 (24%) | 12 (24%) | 9 (18%) |

**TABLE B5**
**Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Alimentary System** (continued) | | | | |
| Liver (continued) | (50) | (50) | (50) | (50) |
| Centrilobular, necrosis | 1 (2%) | | 1 (2%) | |
| Hepatocyte, hyperplasia, focal | | 1 (2%) | | |
| Hepatocyte, vacuolization cytoplasmic | 7 (14%) | 6 (12%) | 2 (4%) | |
| Kupffer cell, hyperplasia | 1 (2%) | 1 (2%) | | |
| Kupffer cell, pigmentation | 4 (8%) | 1 (2%) | 5 (10%) | 10 (20%) |
| Lobules, necrosis | 6 (12%) | 3 (6%) | | 6 (12%) |
| Mesentery | (10) | (8) | (8) | (6) |
| Accessory spleen | 1 (10%) | | 1 (13%) | 1 (17%) |
| Fat, necrosis | 9 (90%) | 7 (88%) | 6 (75%) | 6 (100%) |
| Oral mucosa | | (1) | | (1) |
| Hyperplasia | | | | 1 (100%) |
| Pancreas | (50) | (50) | (50) | (50) |
| Atrophy | 18 (36%) | 18 (36%) | 16 (32%) | 12 (24%) |
| Edema | | 1 (2%) | | |
| Acinus, cytoplasmic alteration | 1 (2%) | 1 (2%) | 1 (2%) | |
| Acinus, hyperplasia, focal | 1 (2%) | 4 (8%) | 2 (4%) | |
| Salivary glands | (49) | (50) | (50) | (50) |
| Atrophy | 1 (2%) | 2 (4%) | | |
| Hyperplasia | 1 (2%) | | | |
| Stomach, forestomach | (50) | (50) | (50) | (50) |
| Edema | 2 (4%) | 1 (2%) | 1 (2%) | |
| Ulcer | | 1 (2%) | | 1 (2%) |
| Mucosa, hyperplasia | 1 (2%) | 2 (4%) | 2 (4%) | 1 (2%) |
| Stomach, glandular | (50) | (50) | (50) | (50) |
| Edema | | | 1 (2%) | |
| Erosion | 1 (2%) | 2 (4%) | | |
| Hyperplasia | | | 1 (2%) | |
| Inflammation, chronic | 1 (2%) | | | |
| Mineralization | 1 (2%) | | | |
| Ulcer | 3 (6%) | 1 (2%) | 1 (2%) | |
| Tongue | (3) | | (1) | |
| Epithelium, hyperplasia | 1 (33%) | | 1 (100%) | |
| **Cardiovascular System** | | | | |
| Heart | (50) | (50) | (50) | (50) |
| Cardiomyopathy | 16 (32%) | 21 (42%) | 23 (46%) | 13 (26%) |
| Mineralization | | | | 1 (2%) |
| Thrombosis | 1 (2%) | | | 1 (2%) |
| **Endocrine System** | | | | |
| Adrenal cortex | (50) | (50) | (50) | (50) |
| Accessory adrenal cortical nodule | 5 (10%) | 6 (12%) | 8 (16%) | 6 (12%) |
| Angiectasis | 22 (44%) | 23 (46%) | 26 (52%) | 29 (58%) |
| Cyst | | | | 1 (2%) |
| Degeneration, fatty | 10 (20%) | 15 (30%) | 11 (22%) | 4 (8%) |
| Hemorrhage | | 1 (2%) | | 1 (2%) |
| Hyperplasia, focal | 9 (18%) | 4 (8%) | 2 (4%) | 2 (4%) |
| Hypertrophy, focal | 9 (18%) | 11 (22%) | 7 (14%) | |
| Necrosis | | | 1 (2%) | |

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| *2-Year Study* (continued) | | | | |
| **Endocrine System** (continued) | | | | |
| Adrenal medulla | (49) | (49) | (50) | (50) |
| Hyperplasia | 1 (2%) | 2 (4%) | 4 (8%) | 1 (2%) |
| Islets, pancreatic | (50) | (50) | (50) | (50) |
| Hyperplasia | | 1 (2%) | | |
| Metaplasia, hepatocyte | | | | 1 (2%) |
| Parathyroid gland | (47) | (49) | (49) | (48) |
| Hyperplasia | | 1 (2%) | | |
| Pituitary gland | (49) | (50) | (48) | (50) |
| Pars distalis, angiectasis | 7 (14%) | 7 (14%) | 8 (17%) | 7 (14%) |
| Pars distalis, cyst | 24 (49%) | 20 (40%) | 16 (33%) | 19 (38%) |
| Pars distalis, hyperplasia | | | | 1 (2%) |
| Pars distalis, hyperplasia, focal | 12 (24%) | 11 (22%) | 9 (19%) | 11 (22%) |
| Pars intermedia, angiectasis | | 3 (6%) | 3 (6%) | |
| Pars intermedia, cyst | | 2 (4%) | | |
| Pars intermedia, hyperplasia | 1 (2%) | 1 (2%) | | |
| Thyroid gland | (49) | (50) | (50) | (50) |
| Ultimobranchial cyst | 2 (4%) | 1 (2%) | 1 (2%) | |
| C-cell, hyperplasia | 6 (12%) | 6 (12%) | 7 (14%) | 3 (6%) |
| Follicle, cyst | | 1 (2%) | 1 (2%) | |
| Follicular cell, hyperplasia | | | | 1 (2%) |
| **General Body System** | | | | |
| Tissue NOS | (1) | | | (1) |
| Hemorrhage | 1 (100%) | | | |
| **Genital System** | | | | |
| Clitoral gland | (50) | (50) | (50) | (50) |
| Ectasia | 12 (24%) | 21 (42%) | 13 (26%) | 10 (20%) |
| Hyperplasia | 1 (2%) | 4 (8%) | 2 (4%) | 1 (2%) |
| Inflammation, chronic | 4 (8%) | 4 (8%) | 3 (6%) | 1 (2%) |
| Inflammation, suppurative | 3 (6%) | 5 (10%) | 6 (12%) | 3 (6%) |
| Ovary | (50) | (50) | (50) | (50) |
| Cyst | 3 (6%) | 5 (10%) | 6 (12%) | 5 (10%) |
| Inflammation, chronic | 1 (2%) | | 1 (2%) | |
| Uterus | (50) | (50) | (50) | (50) |
| Cyst | | | 1 (2%) | |
| Hydrometra | 2 (4%) | 1 (2%) | 3 (6%) | 3 (6%) |
| Hyperplasia, cystic | 4 (8%) | 4 (8%) | 1 (2%) | 11 (22%) |
| **Hematopoietic System** | | | | |
| Bone marrow | (50) | (50) | (50) | (50) |
| Hypercellularity | 1 (2%) | 2 (4%) | | 1 (2%) |
| Myelofibrosis | 4 (8%) | 3 (6%) | 6 (12%) | 6 (12%) |
| Lymph node | (12) | (11) | (16) | (15) |
| Iliac, hyperplasia, lymphoid | | 1 (9%) | | |
| Iliac, pigmentation | | 1 (9%) | | |
| Mediastinal, hemorrhage | 1 (8%) | 1 (9%) | 1 (6%) | 1 (7%) |
| Mediastinal, hyperplasia, lymphoid | | 1 (9%) | | |
| Mediastinal, pigmentation | 2 (17%) | 5 (45%) | 9 (56%) | 9 (60%) |

TABLE B5
**Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| ***2-Year Study*** (continued) | | | | |
| **Hematopoietic System** (continued) | | | | |
| Lymph node (continued) | (12) | (11) | (16) | (15) |
| Pancreatic, hemorrhage | | | 1 (6%) | |
| Pancreatic, pigmentation | 1 (8%) | 2 (18%) | 3 (19%) | 3 (20%) |
| Renal, hemorrhage | | 2 (18%) | 2 (13%) | |
| Renal, hyperplasia, lymphoid | | 1 (9%) | | |
| Renal, pigmentation | 1 (8%) | 5 (45%) | 3 (19%) | 4 (27%) |
| Lymph node, mandibular | (49) | (50) | (50) | (50) |
| Ectasia | 1 (2%) | 4 (8%) | 2 (4%) | 3 (6%) |
| Hemorrhage | 5 (10%) | 9 (18%) | 3 (6%) | 5 (10%) |
| Hyperplasia, lymphoid | 6 (12%) | 6 (12%) | 3 (6%) | |
| Pigmentation | 12 (24%) | 23 (46%) | 13 (26%) | 16 (32%) |
| Lymph node, mesenteric | (50) | (50) | (50) | (50) |
| Ectasia | 1 (2%) | | | |
| Hemorrhage | 3 (6%) | 3 (6%) | 3 (6%) | 2 (4%) |
| Hyperplasia, lymphoid | 1 (2%) | 2 (4%) | 1 (2%) | |
| Spleen | (50) | (50) | (50) | (50) |
| Developmental malformation | | | | 1 (2%) |
| Fibrosis | 1 (2%) | 2 (4%) | 3 (6%) | 1 (2%) |
| Hematopoietic cell proliferation | 10 (20%) | 12 (24%) | 8 (16%) | 14 (28%) |
| Hemorrhage | | 1 (2%) | | |
| Necrosis | 1 (2%) | | | |
| Pigmentation, hemosiderin | 19 (38%) | 20 (40%) | 21 (42%) | 29 (58%) |
| Thymus | (49) | (50) | (50) | (48) |
| Cyst | | 1 (2%) | | |
| Hemorrhage | 1 (2%) | | 1 (2%) | 1 (2%) |
| **Integumentary System** | | | | |
| Mammary gland | (49) | (50) | (50) | (50) |
| Ectasia | 12 (24%) | 9 (18%) | 20 (40%) | 11 (22%) |
| Hyperplasia | 30 (61%) | 39 (78%) | 22 (44%) | 17 (34%) |
| Inflammation, chronic active | | | 1 (2%) | |
| Skin | (50) | (50) | (50) | (50) |
| Acanthosis | 1 (2%) | 2 (4%) | | 1 (2%) |
| Cyst epithelial inclusion | 1 (2%) | | 1 (2%) | |
| Developmental malformation | | | 1 (2%) | |
| Hemorrhage | | | 2 (4%) | |
| Hyperkeratosis | 3 (6%) | 2 (4%) | 3 (6%) | |
| Inflammation, chronic | 1 (2%) | | | |
| Ulcer | 2 (4%) | 4 (8%) | | 2 (4%) |
| **Musculoskeletal System** | | | | |
| Bone | (50) | (50) | (50) | (50) |
| Cranium, osteopetrosis | 7 (14%) | 10 (20%) | 9 (18%) | 6 (12%) |
| Femur, osteopetrosis | 5 (10%) | 6 (12%) | 6 (12%) | 3 (6%) |
| Femur, osteosclerosis | | | 1 (2%) | |
| Rib, osteopetrosis | | | | 1 (2%) |
| Skeletal muscle | (1) | | | (1) |
| Hemorrhage | 1 (100%) | | | 1 (100%) |

TABLE B5
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| *2-Year Study* (continued) | | | | |
| **Nervous System** | | | | |
| Brain | (50) | (50) | (50) | (50) |
| Compression | 9 (18%) | 6 (12%) | 9 (18%) | 8 (16%) |
| Gliosis | 1 (2%) | | | |
| Hemorrhage | | 1 (2%) | 1 (2%) | |
| Hydrocephalus | 2 (4%) | 3 (6%) | | 1 (2%) |
| Necrosis | | 1 (2%) | | |
| **Respiratory System** | | | | |
| Lung | (50) | (50) | (50) | (50) |
| Congestion | | | 1 (2%) | 1 (2%) |
| Hemorrhage | | 2 (4%) | | 1 (2%) |
| Infiltration cellular, histiocyte | 21 (42%) | 22 (44%) | 18 (36%) | 19 (38%) |
| Inflammation, subacute | 1 (2%) | | 2 (4%) | 1 (2%) |
| Alveolar epithelium, hyperplasia | 2 (4%) | | 4 (8%) | 2 (4%) |
| Nose | (50) | (50) | (50) | (50) |
| Exudate | 6 (12%) | 3 (6%) | 2 (4%) | 4 (8%) |
| Foreign body | 5 (10%) | 2 (4%) | 1 (2%) | |
| Fungus | 3 (6%) | 1 (2%) | 1 (2%) | |
| Mucosa, hyperplasia | 3 (6%) | 2 (4%) | 2 (4%) | 1 (2%) |
| Mucosa, metaplasia, squamous | 3 (6%) | | 1 (2%) | 1 (2%) |
| **Special Senses System** | | | | |
| Eye | (2) | | (1) | (3) |
| Atrophy | 1 (50%) | | | |
| Cataract | | | | 2 (67%) |
| Hemorrhage | 1 (50%) | | | 1 (33%) |
| Retina, degeneration | | | | 2 (67%) |
| **Urinary System** | | | | |
| Kidney | (50) | (50) | (50) | (50) |
| Calculus, microscopic observation only | | | | 1 (2%) |
| Cyst | | | 2 (4%) | 2 (4%) |
| Hydronephrosis | | | 1 (2%) | 1 (2%) |
| Mineralization | 43 (86%) | 34 (68%) | 37 (74%) | 35 (70%) |
| Nephropathy | 34 (68%) | 47 (94%) | 43 (86%) | 45 (90%) |
| Renal tubule, atrophy | | 1 (2%) | 3 (6%) | 2 (4%) |
| Renal tubule, cytoplasmic alteration | 1 (2%) | | | |
| Renal tubule, dilatation | 1 (2%) | | 2 (4%) | 1 (2%) |
| Renal tubule, necrosis | 1 (2%) | | | 1 (2%) |
| Renal tubule, pigmentation | 49 (98%) | 49 (98%) | 49 (98%) | 47 (94%) |
| Transitional epithelium, hyperplasia | | 3 (6%) | 7 (14%) | 4 (8%) |
| Ureter | | | (1) | (1) |
| Dilatation | | | | 1 (100%) |
| Transitional epithelium, hyperplasia | | | | 1 (100%) |
| Urinary bladder | (50) | (50) | (50) | (50) |
| Edema | 1 (2%) | | 1 (2%) | |
| Hemorrhage | | | 1 (2%) | |
| Transitional epithelium, hyperplasia | 4 (8%) | | 1 (2%) | 10 (20%) |

# APPENDIX C
# GENETIC TOXICOLOGY

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# GENETIC TOXICOLOGY

## *SALMONELLA TYPHIMURIUM* MUTAGENICITY TEST PROTOCOL

Testing was performed as reported by Zeiger *et al.* (1985). Butyl benzyl phthalate was sent to the laboratory as a coded aliquot from Radian Corporation (Austin, TX). It was incubated with the *Salmonella typhimurium* tester strains (TA98, TA100, TA1535, and TA1537) 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 butyl benzyl phthalate. In the absence of toxicity, 10,000 or 11,500 μg/plate was selected as the high dose. All trials were repeated.

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 was no minimum percentage or fold increase required for a chemical to be judged positive or weakly positive.

## MOUSE LYMPHOMA MUTAGENICITY TEST PROTOCOL

The experimental protocol is presented in detail by Myhr and Caspary (1991). Butyl benzyl phthalate was supplied as a coded aliquot by Radian Corporation. The high dose of butyl benzyl phthalate was determined by toxicity. Mouse lymphoma L5178Y cells were maintained at 37° C as suspension cultures in supplemented Fischer's medium; normal cycling time was approximately 10 hours. To reduce the number of spontaneously occurring trifluorothymidine-resistant cells, subcultures were exposed to medium containing THMG (thymidine, hypoxanthine, methotrexate, and glycine) for 1 day, to medium containing THG (thymidine, hypoxanthine, and glycine) for 1 day, and to normal medium for 3 to 5 days. For cloning, the horse serum content was increased and Noble agar was added.

All treatment levels within an experiment, including concurrent positive and solvent controls, were replicated. Treated cultures contained $6 \times 10^6$ cells in 10 mL medium. This volume included the S9 fraction in those experiments performed with metabolic activation. Incubation with butyl benzyl phthalate continued for 4 hours, at which time the medium plus butyl benzyl phthalate was removed and the cells were resuspended in fresh medium and incubated for an additional 2 days to express the mutant phenotype. Cell density was monitored so that log phase growth was maintained. After the 48-hour expression period, $3 \times 10^6$ cells were plated in medium and soft agar supplemented with trifluorothymidine (TFT) for selection of TFT-resistant ($TK^{-/-}$) cells; 600 cells were plated in nonselective medium and soft agar to determine cloning efficiency. Plates were incubated at 37° C in 5% $CO_2$ for 10 to 12 days. The test was initially performed without S9. If a clearly positive response was not obtained, the test was repeated using freshly prepared S9 from the livers of Aroclor 1254-induced male Fischer 344/N rats.

Minimum criteria for accepting an experiment as valid and a detailed description of the statistical analysis and data evaluation are presented in Caspary *et al.* (1988). All data were evaluated statistically for trend and peak responses. Both responses had to be significant ($P \leq 0.05$) for butyl benzyl phthalate to be

considered positive, i.e., capable of inducing TFT resistance. A single significant response led to a "questionable" conclusion, and the absence of both a trend and peak response resulted in a "negative" call.

## Chinese Hamster Ovary Cell Cytogenetics Protocols

Testing was performed as reported by Galloway *et al.* (1987). Butyl benzyl phthalate 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 butyl benzyl phthalate; the high dose was limited by toxicity to 1,250 μg/mL. A single flask per dose was used, and tests yielding equivocal or positive results were repeated.

***Sister Chromatid Exchange Test:*** In the SCE test without S9, CHO cells were incubated for 26 hours with butyl benzyl phthalate in supplemented McCoy's 5A medium. Bromodeoxyuridine (BrdU) was added 2 hours after culture initiation. After 26 hours, the medium containing butyl benzyl phthalate 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 butyl benzyl phthalate, serum-free medium, and S9 for 2 hours. The medium was then removed and replaced with medium containing serum and BrdU and no butyl benzyl phthalate, 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.

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 ($P \leq 0.05$) 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 butyl benzyl phthalate for 12 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 butyl benzyl phthalate and S9 for 2 hours, after which the treatment medium was removed and the cells were incubated for 12 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.

Cells were selected for scoring on the basis of good morphology and completeness of karyotype ($21 \pm 2$ chromosomes). All slides were scored blind, and those from a single test were read by the same person. One hundred first-division metaphase cells were scored at each dose level. 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 percentage 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.

## *DROSOPHILA MELANOGASTER* TEST PROTOCOL

The assays for induction of sex-linked recessive lethal (SLRL) mutations were performed with adult flies as described by Valencia *et al.* (1985). Butyl benzyl phthalate was supplied as a coded aliquot by Radian Corporation. It was assayed in the SLRL test by feeding for 3 days to adult Canton-S wild-type males no more than 24 hours old at the beginning of treatment. Because no response was obtained, butyl benzyl phthalate was retested by injection into adult males.

To administer butyl benzyl phthalate by injection, a glass Pasteur pipette was drawn out in a flame to a microfine filament, and the tip was broken off to allow delivery of the test solution. Injection was performed either manually, by attaching a rubber bulb to the other end of the pipette and forcing through sufficient solution (0.2 to 0.3 $\mu$L) to slightly distend the abdomen of the fly, or by attaching the pipette to a microinjector which automatically delivered a calibrated volume. Flies were anesthetized with ether and immobilized on a strip of tape. Injection into the thorax, under the wing, was performed with the aid of a dissecting microscope.

Toxicity tests were performed to set concentrations of butyl benzyl phthalate at a level that would induce 30% mortality after 72 hours of feeding or 24 hours after injection, while keeping induced sterility at an acceptable level. Oral exposure was achieved by allowing Canton-S males to feed for 72 hours on a solution of butyl benzyl phthalate in 5% sucrose. In the injection experiments, 24- to 72-hour old Canton-S males were treated with a solution of butyl benzyl phthalate dissolved in 9% ethanol:1% Tween-80 and allowed to recover for 24 hours. A concurrent solvent control group was also included. In the adult exposures, treated males were mated to three *Basc* females for 3 days and given fresh females at 2-day intervals to produce three matings of 3, 2, and 2 days (in each case, sample sperm from successive matings were treated at successively earlier post-meiotic stages). $F_1$ heterozygous females were mated with their siblings and then placed in individual vials. $F_1$ daughters from the same parental male were kept together to identify clusters. (A cluster occurs when a number of mutants from a given male result from a single spontaneous premeiotic mutation event, and is identified when the number of mutants from that male exceeds the number predicted by a Poisson distribution.) If a cluster was identified, all data from the male in question were discarded. Presumptive lethal mutations were identified as vials containing fewer than 5% of the expected number of wild-type males after 17 days; these were retested to confirm the response.

SLRL data were analyzed by simultaneous comparison with the concurrent and historical controls, using a normal approximation to the binomial test (Margolin *et al.*, 1983). A test result was considered positive if $P \leq 0.01$ and the mutation frequency in the tested group was greater than 0.10%, or if $P \leq 0.05$ and the frequency in the treatment group was greater than 0.15%. A test was considered to be inconclusive if (a) the P value was between 0.05 and 0.01 but the frequency in the treatment group was between 0.10% and 0.15% or (b) the P value was between 0.10 and 0.05 but the frequency in the treatment group was greater than 0.10%. A test was considered negative if $P \geq 0.10$ or if the frequency in the treatment group was less than 0.10%.

## MOUSE BONE MARROW SISTER CHROMATID EXCHANGE TEST PROTOCOLS

Doses were selected based on available $LD_{50}$ information, and the high dose was limited by experimental design to 5,000 mg/kg. Butyl benzyl phthalate was tested for the induction of SCEs in mouse bone marrow using two protocols. Male $B6C3F_1$ mice (five animals per dose group) were injected intraperitoneally with butyl benzyl phthalate dissolved in corn oil (injection volume = 0.4 mL). Solvent control mice received equivalent injections of corn oil only. The positive control was dimethylbenzanthracene.

The first protocol had a standard harvest time of 23 hours, and the second protocol had a delayed harvest time of 42 hours. The mice were implanted subcutaneously with a BrdU tablet (McFee *et al.*, 1983) 24 hours before harvest (1 hour before butyl benzyl phthalate treatment in the case of the standard protocol). The use of BrdU allowed selection of the appropriate cell population (cells in the second metaphase following butyl benzyl phthalate treatment) for scoring. Two hours before sacrifice, the mice received an intraperitoneal injection of colchicine in saline. The animals were killed 23 or 42 hours after treatment (24 hours after BrdU dosing). One or both femurs were removed, and the marrow was flushed out with phosphate-buffered saline (pH 7.0). The cells were treated with a hypotonic salt solution, fixed, and dropped onto chilled slides. After a 24-hour drying period, the slides were stained using fluorescence-plus-Giemsa and scored.

Twenty-five second-division metaphase cells were scored from each of four animals per treatment group. Responses were evaluated as SCEs/cell, and the data were analyzed by a trend test (Margolin *et al.*, 1986).

## MOUSE BONE MARROW CHROMOSOMAL ABERRATIONS TEST PROTOCOLS

Doses were selected based on available $LD_{50}$ information, and the high dose was limited by experimental design to 5,000 mg/kg. Butyl benzyl phthalate was tested for induction of Abs in mouse bone marrow using two different protocols. The first protocol used a standard harvest time of 17 hours, and the second protocol used a delayed harvest time of 36 hours.

Male $B6C3F_1$ mice (10 animals per dose group) were injected intraperitoneally with butyl benzyl phthalate dissolved in corn oil (injection volume = 0.4 mL). Solvent control mice received equivalent injections of corn oil only. The positive control was dimethylbenzanthracene (100 mg/kg). The mice were subcutaneously implanted with a BrdU tablet (McFee *et al.*, 1983) 18 hours before the scheduled harvest. (For the standard protocol, this required BrdU implantation to precede injection with butyl benzyl phthalate by 1 hour.) The use of BrdU allowed selection of the appropriate cell population for scoring. (Abs induced by chemical administration are present in maximum number at the first metaphase following treatment; they decline in number during subsequent nuclear divisions due to cell death.) Two hours before sacrifice, the mice received an intraperitoneal injection of colchicine in saline. The animals were killed 17 or 36 hours after butyl benzyl phthalate injection (18 hours after BrdU dosing). One or both femurs were removed, and the marrow was flushed out with phosphate-buffered saline (pH 7.0). Cells were treated with a hypotonic salt solution, fixed, and dropped onto chilled slides. After a 24-hour drying period, the slides were stained and scored.

Fifty first-division metaphase cells were scored from each of eight animals per treatment group. The types of aberrations observed (gaps, breaks, rearrangements, and chromatid versus chromosome) were recorded separately for each animal. The mean total number of aberrations and the mean percentage of cells with aberrations (excluding gaps) were determined for each treatment group. The values for percent cells with aberrations were analyzed by a one-tailed trend test (Margolin *et al.*, 1986), and significance was set at $P=0.025$.

## RESULTS

Butyl benzyl phthalate was not mutagenic in a series of *in vitro* tests, but positive responses were obtained in two *in vivo* studies conducted with male mice.

Butyl benzyl phthalate was tested by two laboratories for mutagenicity in *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1537 with and without Aroclor-induced S9 liver enzymes (Table C1; Zeiger *et al.*, 1985). Negative results were obtained at both laboratories in all strains using concentrations of butyl benzyl phthalate that were the highest permitted by experimental design. *In vitro* studies with mammalian cell systems also gave negative results with and without S9. No induction of trifluorothymidine resistance was obtained in L5178Y mouse lymphoma cells with concentrations of butyl benzyl phthalate that formed stable solutions (Table C2; Myhr and Caspary, 1991). Increases in mutant colonies were observed in the absence of S9 in cultures treated with concentrations that produced precipitation, but such responses were not considered valid by experimental quality control parameters. Therefore, the test was concluded to be negative. No induction of SCEs or Abs was observed in CHO cells treated with butyl benzyl phthalate (Tables C3 and C4; Galloway *et al.*, 1987). In the first SCE trial without S9, the response was considered to be equivocal due to a significant trend (P=0.004) in the absence of a significant increase in SCEs at any one dose level. However, results of the second trial were clearly negative, as was the single trial conducted with S9, and the test results were concluded to be negative overall.

No induction of sex-linked recessive lethal mutations was observed in germ cells of male *Drosophila melanogaster* administered 500 ppm butyl benzyl phthalate by injection or up to 50,000 ppm in feed (Table C5; Valencia *et al.*, 1985).

*In vivo* studies with butyl benzyl phthalate in male mice gave positive results for induction of SCEs (Table C6) and Abs (Table C7) at standard sampling times over a dose range of 1,250 to 5,000 mg/kg. In the SCE test, a single trial with sample time of 23 hours post-injection yielded a positive trend when the highest dose was excluded from the analysis (P=0.0067) because of a reduction in response at the 5,000 mg/kg level. The SCE test conducted with a 42-hour sample time also gave a weakly positive response by trend analysis. Neither trial was repeated. Positive trend analyses were obtained in each of two trials in the Abs test that used the standard harvest time of 17 hours post injection. The only significant dose in each trial was the highest dose (5,000 mg/kg). The single Abs trial that used a delayed harvest time of 36 hours showed no increases in Abs at any of the three dose levels tested.

In conclusion, butyl benzyl phthalate showed no evidence of mutagenicity in any of four *in vitro* tests performed with and without S9 activation enzymes. A test for induction of SLRL mutations in germ cells of male *Drosophila melanogaster* also gave negative results. Positive results were obtained in mouse bone marrow tests for induction of SCEs and Abs. Responses were rather weak, however, and the SCE test was not repeated.

TABLE C1
Mutagenicity of Butyl Benzyl Phthalate in *Salmonella typhimurium*[a]

| Strain | Dose (μg/plate) | Revertants/plate[b] -S9 Trial 1 | -S9 Trial 2 | +10% hamster S9 Trial 1 | +10% hamster S9 Trial 2 | +10% rat S9 Trial 1 | +10% rat S9 Trial 2 |
|---|---|---|---|---|---|---|---|
| **Study performed at EG&G Mason Research, Inc.** | | | | | | | |
| TA100 | 0 | 129 ± 2.7 | 130 ± 4.4 | 113 ± 6.2 | 142 ± 4.4 | 111 ± 10.0 | 141 ± 4.6 |
| | 100 | 140 ± 6.2 | 133 ± 8.4 | 118 ± 2.3 | 123 ± 5.5 | 120 ± 1.2 | 150 ± 13.7 |
| | 333 | 134 ± 7.5 | 142 ± 2.7 | 110 ± 8.9 | 130 ± 4.9 | 118 ± 11.2 | 140 ± 7.2 |
| | 1,000 | 135 ± 4.2 | 146 ± 5.1 | 107 ± 5.0 | 118 ± 9.8 | 117 ± 5.0 | 125 ± 9.2 |
| | 3,333 | 138 ± 4.7 | 137 ± 12.5 | 128 ± 3.3 | 127 ± 2.6 | 109 ± 7.8 | 122 ± 8.7 |
| | 10,000 | 140 ± 3.0 | 136 ± 7.8 | 121 ± 13.5 | 119 ± 15.7 | 102 ± 0.6 | 124 ± 4.1 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control[c] | | 1,539 ± 54.0 | 1,321 ± 82.6 | 1,767 ± 89.4 | 2,031 ± 117.4 | 929 ± 12.3 | 1,191 ± 36.6 |
| TA1535 | 0 | 24 ± 1.5 | 16 ± 0.9 | 14 ± 1.0 | 10 ± 2.1 | 9 ± 2.0 | 12 ± 0.9 |
| | 100 | 19 ± 2.1 | 15 ± 2.1 | 10 ± 3.7 | 9 ± 2.4 | 8 ± 0.9 | 11 ± 1.0 |
| | 333 | 16 ± 0.9 | 16 ± 2.0 | 8 ± 1.5 | 16 ± 1.2 | 9 ± 1.7 | 12 ± 2.3 |
| | 1,000 | 19 ± 1.8 | 17 ± 2.2 | 10 ± 0.6 | 8 ± 2.4 | 6 ± 1.0 | 16 ± 1.3 |
| | 3,333 | 16 ± 1.8 | 13 ± 2.0 | 9 ± 1.5 | 12 ± 4.5 | 4 ± 0.3 | 7 ± 0.7 |
| | 10,000 | 16 ± 0.3 | 14 ± 1.3 | 6 ± 1.8 | 7 ± 0.7 | 7 ± 0.6 | 9 ± 1.2 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 1,041 ± 90.4 | 1,270 ± 48.2 | 146 ± 7.6 | 147 ± 7.8 | 47 ± 4.9 | 57 ± 3.4 |
| TA1537 | 0 | 4 ± 0.0 | 7 ± 1.5 | 8 ± 0.9 | 8 ± 2.2 | 12 ± 1.0 | 7 ± 1.5 |
| | 100 | 5 ± 1.5 | 9 ± 2.6 | 9 ± 0.6 | 10 ± 3.8 | 9 ± 0.7 | 11 ± 0.6 |
| | 333 | 5 ± 0.7 | 6 ± 0.3 | 7 ± 0.6 | 8 ± 3.3 | 10 ± 1.5 | 6 ± 2.5 |
| | 1,000 | 5 ± 1.0 | 11 ± 1.3 | 9 ± 1.0 | 7 ± 0.3 | 11 ± 3.3 | 11 ± 2.2 |
| | 3,333 | 7 ± 1.2 | 7 ± 0.7 | 8 ± 2.4 | 4 ± 0.6 | 11 ± 2.1 | 8 ± 1.5 |
| | 10,000 | 8 ± 1.7 | 7 ± 0.9 | 7 ± 0.6 | 5 ± 1.2 | 3 ± 0.3 | 6 ± 1.2 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 265 ± 30.6 | 466 ± 30.3 | 140 ± 5.0 | 222 ± 24.1 | 58 ± 4.4 | 112 ± 8.7 |
| TA98 | 0 | 23 ± 1.0 | 22 ± 0.9 | 32 ± 0.9 | 32 ± 3.7 | 32 ± 1.5 | 27 ± 3.8 |
| | 100 | 19 ± 2.1 | 26 ± 4.3 | 30 ± 2.3 | 30 ± 2.0 | 27 ± 0.6 | 36 ± 0.9 |
| | 333 | 21 ± 1.9 | 27 ± 3.4 | 23 ± 5.7 | 27 ± 1.5 | 28 ± 4.2 | 30 ± 4.0 |
| | 1,000 | 15 ± 2.7 | 16 ± 0.9 | 26 ± 1.5 | 27 ± 1.2 | 31 ± 3.1 | 28 ± 4.3 |
| | 3,333 | 19 ± 2.2 | 21 ± 5.4 | 27 ± 1.8 | 23 ± 2.3 | 20 ± 4.9 | 25 ± 2.5 |
| | 10,000 | 22 ± 2.9 | 21 ± 4.0 | 19 ± 2.9 | 17 ± 2.4 | 20 ± 1.8 | 20 ± 1.2 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 1,624 ± 52.5 | 1,459 ± 51.8 | 1,569 ± 74.6 | 1,928 ± 50.6 | 762 ± 37.1 | 1,056 ± 30.3 |

TABLE C1
**Mutagenicity of Butyl Benzyl Phthalate in *Salmonella typhimurium*** (continued)

| Strain | Dose (μg/plate) | Revertants/plate -S9 Trial 1 | -S9 Trial 2 | +10% hamster S9 Trial 1 | +10% hamster S9 Trial 2 | +10% rat S9 Trial 1 | +10% rat S9 Trial 2 |
|---|---|---|---|---|---|---|---|
| **Study performed at Case Western Reserve University** | | | | | | | |
| **TA100** | 0 | 99 ± 1.2 | 108 ± 9.8 | 160 ± 8.6 | 169 ± 8.0 | 164 ± 15.7 | 142 ± 11.7 |
| | 333 | 104 ± 2.9 | 117 ± 1.8 | 180 ± 14.5 | 176 ± 10.2 | 162 ± 2.2 | 143 ± 3.0 |
| | 1,000 | 109 ± 0.9 | 117 ± 4.2 | 162 ± 3.5 | 169 ± 2.2 | 138 ± 11.4 | 145 ± 13.3 |
| | 3,333 | 123 ± 11.2 | 125 ± 5.5 | 156 ± 2.9 | 149 ± 8.8 | 157 ± 3.0 | 159 ± 12.3 |
| | 10,000 | 125 ± 2.2 | 121 ± 9.9 | 222 ± 4.6 | 180 ± 17.0 | 171 ± 13.5 | 170 ± 12.2 |
| | 11,550 | 155 ± 2.9 | 132 ± 19.3 | 196 ± 10.0 | 178 ± 18.4 | 186 ± 4.0 | 163 ± 1.2 |
| Trial summary | | Equivocal | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 733 ± 35.0 | 722 ± 23.9 | 2,057 ± 90.3 | 1,023 ± 28.3 | 2,440 ± 73.1 | 1,359 ± 165.2 |
| **TA1535** | 0 | 5 ± 1.0 | 8 ± 0.3 | 6 ± 1.2 | 13 ± 2.0 | 8 ± 1.8 | 13 ± 0.9 |
| | 333 | 3 ± 0.3 | 6 ± 0.6 | 8 ± 0.7 | 12 ± 4.4 | 5 ± 0.7 | 8 ± 0.3 |
| | 1,000 | 2 ± 0.9 | 4 ± 0.9 | 5 ± 0.7 | 9 ± 1.0 | 7 ± 1.5 | 9 ± 0.9 |
| | 3,333 | 4 ± 0.0 | 7 ± 0.7 | 5 ± 1.8 | 10 ± 0.3 | 8 ± 0.9 | 10 ± 2.1 |
| | 10,000 | 3 ± 0.3 | 7 ± 1.7 | 5 ± 1.0 | 8 ± 1.5 | 4 ± 1.0 | 9 ± 2.1 |
| | 11,550 | 6 ± 0.9 | 9 ± 1.5 | 6 ± 1.0 | 10 ± 1.8 | 5 ± 1.8 | 8 ± 1.5 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 435 ± 50.6 | 957 ± 173.9 | 93 ± 9.3 | 117 ± 4.6 | 160 ± 13.5 | 182 ± 15.7 |
| **TA1537** | 0 | 2 ± 0.6 | 5 ± 2.3 | 8 ± 0.6 | 10 ± 2.3 | 7 ± 1.5 | 9 ± 1.2 |
| | 333 | 3 ± 1.2 | 5 ± 1.7 | 7 ± 0.3 | 9 ± 0.3 | 5 ± 0.3 | 8 ± 2.3 |
| | 1,000 | 5 ± 1.0 | 6 ± 3.2 | 5 ± 0.3 | 8 ± 1.2 | 7 ± 0.3 | 7 ± 2.3 |
| | 3,333 | 3 ± 0.7 | 7 ± 0.6 | 5 ± 0.0 | 10 ± 2.0 | 6 ± 0.9 | 9 ± 1.9 |
| | 10,000 | 3 ± 0.9 | 5 ± 1.2 | 4 ± 0.0 | 7 ± 2.2 | 8 ± 1.5 | 7 ± 2.6 |
| | 11,550 | 2 ± 0.3 | 6 ± 1.3 | 8 ± 1.5 | 10 ± 2.0 | 7 ± 0.3 | 8 ± 0.9 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 787 ± 185.8 | 507 ± 94.8 | 122 ± 5.7 | 71 ± 3.8 | 117 ± 30.4 | 122 ± 19.9 |
| **TA98** | 0 | 12 ± 2.3 | 22 ± 6.2 | 19 ± 1.8 | 25 ± 3.6 | 21 ± 5.7 | 22 ± 1.5 |
| | 333 | 8 ± 0.7 | 13 ± 0.7 | 19 ± 1.0 | 28 ± 4.3 | 23 ± 1.9 | 21 ± 2.1 |
| | 1,000 | 11 ± 0.7 | 13 ± 3.8 | 21 ± 2.4 | 27 ± 2.3 | 23 ± 1.7 | 24 ± 2.6 |
| | 3,333 | 11 ± 0.6 | 14 ± 3.1 | 18 ± 4.4 | 26 ± 2.2 | 25 ± 2.0 | 23 ± 1.8 |
| | 10,000 | 13 ± 0.9 | 17 ± 3.7 | 14 ± 1.0 | 21 ± 1.5 | 21 ± 2.5 | 26 ± 3.8 |
| | 11,550 | 11 ± 2.1 | 17 ± 2.0 | 25 ± 3.0 | 28 ± 4.0 | 29 ± 2.3 | 29 ± 0.6 |
| Trial summary | | Negative | Negative | Negative | Negative | Negative | Negative |
| Positive control | | 195 ± 27.9 | 283 ± 19.2 | 1,275 ± 368.8 | 542 ± 28.1 | 1,454 ± 115.4 | 622 ± 110.8 |

[a] The detailed protocol and these data are presented in Zeiger *et al.* (1985).
[b] Revertants are presented as mean ± standard error from three plates.
[c] The positive controls in the absence of metabolic activation were sodium azide (TA1535 and TA100), 9-aminoacridine (TA1537), and 4-nitro-*o*-phenylenediamine (TA98). The positive control for metabolic activation with all strains was 2-aminoanthracene.

TABLE C2
Induction of Trifluorothymidine Resistance in L5178Y Mouse Lymphoma Cells by Butyl Benzyl Phthalate[a]

| Compound | Concentration (nL/mL) | Cloning Efficiency (%) | Relative Total Growth (%) | Mutant Count | Mutant Fraction[b] | Average Mutant Fraction[c] |
|---|---|---|---|---|---|---|
| -S9 | | | | | | |
| Trial 1 | | | | | | |
| Ethanol | | 77 | 72 | 85 | 37 | |
| | | 94 | 122 | 108 | 38 | |
| | | 74 | 92 | 67 | 30 | |
| | | 114 | 114 | 96 | 28 | 33 |
| Methyl methanesulfonate | 5[d] | 62 | 37 | 606 | 325 | |
| | | 33 | 18 | 491 | 501 | |
| | | 57 | 33 | 556 | 327 | 384[e] |
| Butyl benzyl phthalate | 5 | 74 | 61 | 87 | 39 | |
| | | 69 | 82 | 94 | 45 | |
| | | 72 | 77 | 74 | 34 | 40 |
| | 10 | 92 | 72 | 98 | 36 | |
| | | 78 | 68 | 101 | 43 | |
| | | 73 | 76 | 92 | 42 | 40 |
| | 20 | 79 | 40 | 95 | 40 | |
| | | 71 | 34 | 90 | 42 | |
| | | 90 | 38 | 152 | 56 | 46 |
| | 30 | 63 | 14 | 120 | 63 | |
| | | 52 | 23 | 141 | 91 | 77[e] |
| | 40 | 54 | 18 | 173 | 107 | |
| | | 71 | 28 | 132 | 62 | |
| | | 50 | 23 | 126 | 84 | 85[e] |
| | 60[f] | 59 | 5 | 176 | 99 | |
| | | 70 | 10 | 217 | 104 | |
| | | 55 | 7 | 191 | 116 | 106[e] |

TABLE C2
**Induction of Trifluorothymidine Resistance in L5178Y Mouse Lymphoma Cells by Butyl Benzyl Phthalate**
(continued)

| Compound | Concentration (nL/mL) | Cloning Efficiency (%) | Relative Total Growth (%) | Mutant Count | Mutant Fraction | Average Mutant Fraction |
|---|---|---|---|---|---|---|
| **-S9** (continued) | | | | | | |
| **Trial 2** | | | | | | |
| Ethanol | | 84 | 130 | 80 | 32 | |
| | | 93 | 130 | 105 | 38 | |
| | | 82 | 40 | 170 | 69 | 46 |
| | | Toxic | | | | |
| Methyl methanesulfonate | 5[d] | 37 | 12 | 730 | 658 | |
| | | 95 | 101 | 724 | 255 | 456[e] |
| Butyl benzyl phthalate | 20 | 57 | 16 | 167 | 98 | |
| | | 58 | 40 | 63 | 36 | |
| | | 57 | 48 | 80 | 47 | 60 |
| | 30 | 75 | 56 | 110 | 49 | |
| | | 66 | 45 | 106 | 53 | |
| | | 91 | 34 | 128 | 47 | 50 |
| | 40 | 69 | 25 | 99 | 48 | |
| | | 77 | 28 | 153 | 67 | |
| | | 71 | 29 | 125 | 59 | 58 |
| | 50 | 67 | 31 | 95 | 47 | |
| | | 65 | 26 | 99 | 51 | 49 |
| | | Toxic | | | | |
| | 60[f] | 49 | 20 | 115 | 79 | |
| | | 54 | 23 | 97 | 60 | 69 |
| | | Toxic | | | | |
| | 80 | 67 | 18 | 132 | 66 | |
| | | 39 | 15 | 103 | 88 | |
| | | 73 | 27 | 183 | 84 | 79[e] |

TABLE C2
**Induction of Trifluorothymidine Resistance in L5178Y Mouse Lymphoma Cells by Butyl Benzyl Phthalate**
(continued)

| Compound | Concentration (nL/mL) | Cloning Efficiency (%) | Relative Total Growth (%) | Mutant Count | Mutant Fraction | Average Mutant Fraction |
|---|---|---|---|---|---|---|
| -S9 (continued) | | | | | | |
| **Trial 3** | | | | | | |
| Ethanol | | 67 | 79 | 82 | 41 | |
| | | 83 | 110 | 66 | 27 | |
| | | 88 | 117 | 70 | 26 | |
| | | 92 | 94 | 80 | 29 | 31 |
| Methyl methanesulfonate | 5[d] | 58 | 26 | 432 | 248 | |
| | | 49 | 30 | 567 | 384 | |
| | | 54 | 32 | 638 | 398 | 343[e] |
| Butyl benzyl phthalate | 10 | 61 | 69 | 60 | 33 | |
| | | 79 | 72 | 90 | 38 | 35 |
| | 20 | 79 | 61 | 78 | 33 | |
| | | 94 | 39 | 110 | 39 | 36 |
| | 30 | 86 | 39 | 127 | 49 | |
| | | 90 | 27 | 114 | 42 | |
| | | 67 | 36 | 86 | 43 | 45 |
| | 40 | 54 | 24 | 79 | 49 | |
| | | 65 | 17 | 68 | 35 | |
| | | 95 | 43 | 110 | 39 | 41 |
| | 60[f] | 101 | 32 | 135 | 45 | |
| | | 95 | 33 | 115 | 40 | |
| | | 99 | 30 | 102 | 34 | 40 |
| | 80 | 60 | 11 | 94 | 52 | |
| | | 68 | 14 | 106 | 52 | 52[e] |

**TABLE C2**
**Induction of Trifluorothymidine Resistance in L5178Y Mouse Lymphoma Cells by Butyl Benzyl Phthalate**
(continued)

| Compound | Concentration (nL/mL) | Cloning Efficiency (%) | Relative Total Growth (%) | Mutant Count | Mutant Fraction | Average Mutant Fraction |
|---|---|---|---|---|---|---|
| **-S9** (continued) | | | | | | |
| **Trial 4** | | | | | | |
| Ethanol | | 105 | 109 | 62 | 20 | |
| | | 89 | 80 | 68 | 26 | |
| | | 95 | 99 | 70 | 24 | |
| | | 99 | 113 | 82 | 28 | 24 |
| Methyl methanesulfonate | 5[d] | 76 | 73 | 617 | 271 | |
| | | 88 | 93 | 780 | 295 | |
| | | 83 | 69 | 791 | 318 | 295[e] |
| Butyl benzyl phthalate | 10 | 73 | 48 | 35 | 16 | |
| | | 89 | 66 | 46 | 17 | |
| | | 79 | 72 | 49 | 21 | 18 |
| | 20 | 72 | 53 | 41 | 19 | |
| | | 87 | 64 | 54 | 21 | |
| | | 76 | 50 | 53 | 23 | 21 |
| | 30 | 95 | 22 | 49 | 17 | |
| | | 82 | 20 | 61 | 25 | |
| | | 92 | 20 | 57 | 21 | 21 |
| | 40 | Toxic | | | | |
| | | Toxic | | | | |

TABLE C2
Induction of Trifluorothymidine Resistance in L5178Y Mouse Lymphoma Cells by Butyl Benzyl Phthalate
(continued)

| Compound | Concentration (nL/mL) | Cloning Efficiency (%) | Relative Total Growth (%) | Mutant Count | Mutant Fraction | Average Mutant Fraction |
|---|---|---|---|---|---|---|
| +S9 | | | | | | |
| Trial 1 | | | | | | |
| Ethanol | | 104 | 103 | 195 | 62 | |
| | | 82 | 76 | 222 | 91 | |
| | | 80 | 69 | 138 | 57 | |
| | | 104 | 153 | 164 | 52 | 66 |
| Methyl cholanthrene | 2.5[d] | 49 | 27 | 768 | 521 | |
| | | 41 | 19 | 683 | 562 | |
| | | 41 | 10 | 693 | 568 | 550[e] |
| Butyl benzyl phthalate | 30 | 80 | 76 | 150 | 63 | |
| | | 77 | 63 | 203 | 88 | |
| | | 80 | 86 | 158 | 66 | 72 |
| | 40 | 78 | 73 | 116 | 50 | |
| | | 78 | 59 | 149 | 64 | |
| | | 80 | 113 | 104 | 44 | 52 |
| | 50 | 83 | 82 | 128 | 52 | |
| | | 75 | 50 | 123 | 55 | |
| | | 79 | 65 | 101 | 42 | 50 |
| | 60[f] | 96 | 52 | 206 | 71 | |
| | | 81 | 67 | 198 | 81 | |
| | | 74 | 58 | 165 | 75 | 76 |
| | 80 | 78 | 51 | 195 | 84 | |
| | | 76 | 55 | 169 | 74 | |
| | | 80 | 62 | 184 | 77 | 78 |
| | 100 | 79 | 58 | 190 | 80 | |
| | | 73 | 53 | 147 | 67 | |
| | | 79 | 47 | 186 | 78 | 75 |

TABLE C2
**Induction of Trifluorothymidine Resistance in L5178Y Mouse Lymphoma Cells by Butyl Benzyl Phthalate**
(continued)

| Compound | Concentration (nL/mL) | Cloning Efficiency (%) | Relative Total Growth (%) | Mutant Count | Mutant Fraction | Average Mutant Fraction |
|---|---|---|---|---|---|---|
| +S9 (continued) | | | | | | |
| **Trial 2** | | | | | | |
| Ethanol | | 75 | 106 | 112 | 50 | |
| | | 86 | 104 | 73 | 28 | |
| | | 90 | 101 | 107 | 40 | |
| | | 78 | 89 | 104 | 45 | 41 |
| Methyl cholanthrene | 2.5[d] | 69 | 50 | 602 | 291 | |
| | | 76 | 58 | 631 | 277 | |
| | | 64 | 48 | 687 | 359 | 309[e] |
| Butyl benzyl phthalate | 30 | 64 | 65 | 95 | 50 | |
| | | 60 | 59 | 76 | 42 | |
| | | 74 | 64 | 69 | 31 | 41 |
| | 40 | 94 | 81 | 94 | 33 | |
| | | 86 | 57 | 137 | 53 | |
| | | 83 | 66 | 77 | 31 | 39 |
| | 50 | 93 | 59 | 103 | 37 | |
| | | 96 | 67 | 122 | 42 | |
| | | 85 | 77 | 94 | 37 | 39 |
| | 60[f] | 82 | 54 | 92 | 38 | |
| | | 93 | 73 | 111 | 40 | |
| | | 71 | 68 | 89 | 42 | 40 |
| | 80 | 99 | 61 | 121 | 41 | |
| | | 83 | 69 | 90 | 36 | |
| | | 92 | 80 | 109 | 39 | 39 |
| | 100 | 92 | 42 | 99 | 36 | |
| | | 102 | 60 | 98 | 32 | |
| | | 88 | 45 | 96 | 36 | 35 |

[a] Study performed at Litton Bionetics, Inc. The detailed protocol and these data are presented in Myhr and Caspary (1991).
[b] Mutant fraction (MF) (frequency) is a ratio of the mutant count to the cloning efficiency, divided by 3 (to arrive at MF/$10^6$ cells treated).
[c] Mean from three replicate plates of approximately $10^6$ cells each
[d] Chemical was used as a positive control, and the dose is presented as μg/mL.
[e] Significant positive response ($P \leq 0.05$)
[f] Precipitate formed at this dose level.

TABLE C3
Induction of Sister Chromatid Exchanges in Chinese Hamster Ovary Cells by Butyl Benzyl Phthalate[a]

| Compound | Dose (μg/mL) | Total Cells | No. of Chromosomes | No. of SCEs | SCEs/ Chromosome | SCEs/ Cell | Hrs in BrdU | Relative Change of SCEs/ Chromosome[b] (%) |
|---|---|---|---|---|---|---|---|---|
| **-S9** | | | | | | | | |
| **Trial 1** | | | | | | | | |
| Summary: Equivocal | | | | | | | | |
| Dimethylsulfoxide | | 50 | 1,045 | 480 | 0.45 | 9.6 | 26.0 | |
| Triethylenemelamine | 0.015 | 50 | 1,045 | 1,596 | 1.52 | 31.9 | 26.0 | 232.50 |
| Butyl benzyl phthalate | 0.40 | 50 | 1,047 | 549 | 0.52 | 11.0 | 26.0 | 14.16 |
| | 1.25 | 50 | 1,046 | 568 | 0.54 | 11.4 | 26.0 | 18.22 |
| | 4.00 | 50 | 1,046 | 566 | 0.54 | 11.3 | 26.0 | 17.80 |
| | | | | | P=0.004[c] | | | |
| **Trial 2** | | | | | | | | |
| Summary: Negative | | | | | | | | |
| Dimethylsulfoxide | | 50 | 1,043 | 460 | 0.44 | 9.2 | 26.0 | |
| Triethylenemelamine | 0.015 | 50 | 1,048 | 1,482 | 1.41 | 29.6 | 26.0 | 220.64 |
| Butyl benzyl phthalate | 0.40 | 50 | 1,046 | 481 | 0.45 | 9.6 | 26.0 | 4.27 |
| | 1.25 | 50 | 1,052 | 469 | 0.44 | 9.4 | 26.0 | 1.08 |
| | 4.00 | 50 | 1,048 | 405 | 0.38 | 8.1 | 26.0 | -12.38 |
| | 12.50 | 25 | 523 | 219 | 0.41 | 8.8 | 26.0 | -5.06 |
| | 12.50 | 25 | 519 | 230 | 0.44 | 9.2 | 28.0 | 0.48 |
| | | | | | P=0.934 | | | |
| **+S9** | | | | | | | | |
| Summary: Negative | | | | | | | | |
| Dimethylsulfoxide | | 50 | 1,042 | 420 | 0.40 | 8.4 | 26.0 | |
| Cyclophosphamide | 1 | 50 | 1,036 | 1,711 | 1.65 | 34.2 | 26.0 | 309.74 |
| Butyl benzyl phthalate | 125 | 50 | 1,034 | 466 | 0.45 | 9.3 | 26.0 | 11.81 |
| | 400 | 50 | 1,040 | 466 | 0.44 | 9.3 | 26.0 | 11.16 |
| | 1,250 | 50 | 1,039 | 410 | 0.39 | 8.2 | 26.0 | -2.10 |
| | | | | | P=0.618 | | | |

[a] Study performed at Columbia University. The detailed protocol and these data are 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] Significance of SCEs/chromosome tested by the linear regression trend test vs. log of the dose

TABLE C4
**Induction of Chromosomal Aberrations in Chinese Hamster Ovary Cells by Butyl Benzyl Phthalate[a]**

| -S9 | | | | | +S9 | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Dose (μg/mL) | Total Cells Scored | No. of Abs | Abs/ Cell | Cells with Abs (%) | Dose (μg/mL) | Total Cells Scored | No. of Abs | Abs/ Cell | Cells with Abs (%) |
| Harvest time: 14.0 hours | | | | | Harvest time: 14.0 hours | | | | |
| Summary: Negative | | | | | Summary: Negative | | | | |
| Dimethylsulfoxide | | | | | Dimethylsulfoxide | | | | |
| | 100 | 1 | 0.01 | 1.0 | | 100 | 1 | 0.01 | 1.0 |
| Triethylenemelamine | | | | | Cyclophosphamide | | | | |
| 0.15 | 100 | 31 | 0.31 | 25.0 | 15 | 100 | 54 | 0.54 | 35.0 |
| Butyl benzyl phthalate | | | | | Butyl benzyl phthalate | | | | |
| 125 | 100 | 1 | 0.01 | 1.0 | 125 | 100 | 2 | 0.02 | 2.0 |
| 400 | 100 | 2 | 0.02 | 2.0 | 400 | 100 | 3 | 0.03 | 3.0 |
| 1,250 | 100 | 1 | 0.01 | 1.0 | 1,250 | 100 | 1 | 0.01 | 1.0 |
| | | | | P=0.419[b] | | | | | P=0.431 |

[a] Study performed at Columbia University. The detailed protocol and these data are presented in Galloway *et al.* (1987). Abs=aberrations.
[b] Significance of percent cells with aberrations tested by the linear regression trend test vs. log of the dose

TABLE C5
**Induction of Sex-Linked Recessive Lethal Mutations in *Drosophila melanogaster* by Butyl Benzyl Phthalate[a]**

| Route of Exposure | Dose (ppm) | Incidence of Death (%) | No. of Lethals/No. of X Chromosomes Tested | | | Total[b] |
|---|---|---|---|---|---|---|
| | | | Mating 1 | Mating 2 | Mating 3 | |
| Feed | 0 | | 0/1,148 | 0/1,126 | 0/1,019 | 0/3,293 (0.00%)[c] |
| | 10,000 | 40 | 1/1,122 | 1/1,090 | 3/1,107 | 5/3,319 (0.15%) |
| Feed | 0 | | 3/1,058 | 1/1,038 | 2/992 | 6/3,088 (0.19%)[c] |
| | 50,000 | 10 | 0/828 | 0/835 | 1/692 | 1/2,355 (0.04%) |
| Injection | 0 | | 2/2,071 | 3/1,897 | 1/1,581 | 6/5,549 (0.11%) |
| | 500 | 10 | 1/2,007 | 2/1,930 | 2/1,933 | 5/5,870 (0.09%) |

[a] Study performed at University of Wisconsin, Madison. The detailed protocol and these data are presented in Valencia *et al.* (1985). After 17 days, presumptive lethal mutations were identified as vials containing no wild-type males; the parental flies were retested. Results were not significant at the 5% level (Margolin *et al.*, 1983).
[b] Combined total number of lethal mutations/number of X chromosomes tested for three mating trials
[c] P=0.419 for pooled data from both feed studies.

TABLE C6
Induction of Sister Chromatid Exchanges in Mouse Bone Marrow Cells by Butyl Benzyl Phthalate[a]

| Compound | Dose (mg/kg) | Mean SCEs/Cell |
|---|---|---|
| **Trial 1** | | |
| Harvest time: 23 hours | | |
| Corn oil[b] | | 5.98 ± 0.44 |
| Dimethylbenzanthracene[c] | 2.5 | 9.38 ± 0.32 |
| Butyl benzyl phthalate | 1,250 | 6.60 ± 0.85 |
| | 2,500 | 9.22 ± 0.86 |
| | 5,000 | 6.99 ± 0.74 |
| | | P=0.197[d] |
| **Trial 2** | | |
| Harvest time: 42 hours | | |
| Corn oil | | 4.97 ± 0.21 |
| Dimethylbenzanthracene | 2.5 | 12.69 ± 0.54 |
| Butyl benzyl phthalate | 1,250 | 5.75 ± 0.20 |
| | 2,500 | 4.56 ± 0.26 |
| | 5,000 | 5.99 ± 0.21 |
| | | P=0.025 |

[a] Study performed at Brookhaven National Laboratories. SCE=sister chromatid exchange. Twenty-five first-division metaphase cells were scored from each of four animals per treatment group.
[b] Solvent control
[c] Positive control
[d] One-tailed trend analysis (Margolin *et al.*, 1986); significant at P=0.025. For the 23-hour sample time, exclusion of the 5,000 mg/kg dose and recalculation of the trend resulted in a P value of 0.0067, which is significant.

TABLE C7
**Induction of Chromosomal Aberrations in Mouse Bone Marrow Cells by Butyl Benzyl Phthalate[a]**

| Dose (mg/kg) | Total Cells | No. of Abs | Abs/ Cell | Cells with Abs (%) |
|---|---|---|---|---|
| **Trial 1** - Harvest time: 17 hours | | | | |
| Corn oil[b] | | | | |
| | 400 | 2.3 | 0.01 | 0.75 |
| Dimethylbenzanthracene[c] | | | | |
| 100 | 400 | 11.6 | 0.13 | 11.50 |
| Butyl benzyl phthalate | | | | |
| 1,250 | 400 | 2.4 | 0.02 | 1.50 |
| 2,500 | 400 | 0.8 | 0.01 | 0.75 |
| 5,000 | 400 | 3.4 | 0.04 | 3.25* |
| | | | | P=0.003[d] |
| **Trial 2** - Harvest time: 17 hours | | | | |
| Corn oil | | | | |
| | 400 | 1.6 | 0.01 | 1.00 |
| Dimethylbenzanthracene | | | | |
| 100 | 400 | 16.4 | 0.18 | 14.00 |
| Butyl benzyl phthalate | | | | |
| 2,500 | 400 | 4.0 | 0.02 | 2.25 |
| 3,750 | 400 | 2.3 | 0.02 | 2.00 |
| 5,000 | 400 | 6.6 | 0.05 | 4.25** |
| | | | | P=0.003 |
| **Trial 3** - Harvest time: 36 hours | | | | |
| Corn oil | | | | |
| | 400 | 0.5 | 0.0 | 0.25 |
| Dimethylbenzanthracene | | | | |
| 100 | 400 | 56.1 | 0.89 | 26.75 |
| Butyl benzyl phthalate | | | | |
| 1,250 | 400 | 1.0 | 0.01 | 1.50 |
| 2,500 | 400 | 0.4 | 0.00 | 0.25 |
| 5,000 | 400 | 0.5 | 0.01 | 0.50 |
| | | | | P=0.464 |

* Positive (P=0.006)
** P=0.002
[a] Study performed at Brookhaven National Laboratories. Abs=aberrations. Fifty first-division metaphase cells were scored from each of eight animals per treatment group.
[b] Solvent control
[c] Positive control
[d] One-tailed trend analysis (Margolin *et al.*, 1986); significant at P=0.025

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


TABLE D1 Organ Weights and Organ-Weight-to-Body-Weight Ratios for Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate . . . . . 156

TABLE D2 Organ Weights and Organ-Weight-to-Body-Weight Ratios for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate . . . . . 157

TABLE D3 Organ Weights and Organ-Weight-to-Body-Weight Ratios for Rats at the 15-Month Interim Evaluation in the 2-Year Feed Study of Butyl Benzyl Phthalate . . . . . 158

TABLE D1
**Organ Weights and Organ-Weight-to-Body-Weight Ratios for Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate[a]**

| | 0 ppm | 300 ppm | 2,800 ppm | 25,000 ppm |
|---|---|---|---|---|
| n | 15 | 15 | 15 | 15 |
| Necropsy body wt | 317 ± 4 | 315 ± 4 | 310 ± 4 | 225 ± 5** |
| Brain | | | | |
| Absolute | 1.924 ± 0.017 | 1.925 ± 0.017 | 1.903 ± 0.013 | 1.880 ± 0.019 |
| Relative | 6.08 ± 0.07 | 6.12 ± 0.08 | 6.15 ± 0.08 | 8.38 ± 0.13** |
| Heart | | | | |
| Absolute | 1.067 ± 0.032 | 1.032 ± 0.026 | 1.057 ± 0.026 | 0.845 ± 0.027** |
| Relative | 3.37 ± 0.09 | 3.27 ± 0.07 | 3.41 ± 0.09 | 3.75 ± 0.11** |
| R. Kidney | | | | |
| Absolute | 1.231 ± 0.028 | 1.206 ± 0.026 | 1.269 ± 0.029 | 0.926 ± 0.023** |
| Relative | 3.89 ± 0.07 | 3.84 ± 0.11 | 4.09 ± 0.08 | 4.11 ± 0.07 |
| Liver | | | | |
| Absolute | 13.207 ± 0.264 | 13.394 ± 0.230 | 13.170 ± 0.230 | 9.987 ± 0.276** |
| Relative | 41.67 ± 0.51 | 42.51 ± 0.63 | 42.44 ± 0.44 | 44.28 ± 0.62** |
| Lung | | | | |
| Absolute | 1.407 ± 0.036 | 1.473 ± 0.040 | 1.423 ± 0.040 | 1.141 ± 0.036** |
| Relative | 4.45 ± 0.12 | 4.68 ± 0.14 | 4.60 ± 0.15 | 5.07 ± 0.15** |
| Prostate Gland | | | | |
| Absolute | 0.609 ± 0.058 | 0.614 ± 0.061 | 0.604 ± 0.050 | 0.276 ± 0.034** |
| Relative | 1.93 ± 0.19 | 1.95 ± 0.20 | 1.94 ± 0.15 | 1.23 ± 0.15* |
| Seminal Vesicle | | | | |
| Absolute | 1.517 ± 0.081 | 1.290 ± 0.081 | 1.333 ± 0.078 | 1.035 ± 0.059** |
| Relative | 4.80 ± 0.27 | 4.09 ± 0.25 | 4.29 ± 0.24 | 4.58 ± 0.22 |
| R. Testis | | | | |
| Absolute | 1.497 ± 0.017 | 1.378 ± 0.081 | 1.502 ± 0.019 | 0.442 ± 0.014** |
| Relative | 4.73 ± 0.06 | 4.36 ± 0.25 | 4.85 ± 0.06 | 1.97 ± 0.06** |
| Thymus | | | | |
| Absolute | 0.277 ± 0.012 | 0.293 ± 0.017 | 0.271 ± 0.009 | 0.239 ± 0.010 |
| Relative | 0.87 ± 0.04 | 0.92 ± 0.05 | 0.87 ± 0.03 | 1.07 ± 0.05** |

* 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).

TABLE D2
Organ Weights and Organ-Weight-to-Body-Weight Ratios for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 300 ppm | 900 ppm | 2,800 ppm | 8,300 ppm | 25,000 ppm |
|---|---|---|---|---|---|---|
| n | 12 | 14 | 14 | 14 | 15 | 11 |
| Necropsy body wt | 367 ± 16 | 386 ± 5 | 400 ± 6 | 369 ± 8 | 376 ± 7 | 249 ± 13** |
| Brain | | | | | | |
| Absolute | 1.966 ± 0.031 | 2.068 ± 0.018* | 2.081 ± 0.012** | 2.030 ± 0.023 | 2.033 ± 0.018 | 1.916 ± 0.040 |
| Relative | 5.46 ± 0.22 | 5.37 ± 0.08 | 5.22 ± 0.07 | 5.53 ± 0.09 | 5.43 ± 0.11 | 7.98 ± 0.52** |
| Heart | | | | | | |
| Absolute | 1.182 ± 0.066 | 1.179 ± 0.029 | 1.226 ± 0.039 | 1.139 ± 0.034 | 1.140 ± 0.036 | 0.796 ± 0.035** |
| Relative | 3.24 ± 0.14 | 3.06 ± 0.09 | 3.07 ± 0.10 | 3.10 ± 0.09 | 3.03 ± 0.08 | 3.24 ± 0.09 |
| R. Kidney | | | | | | |
| Absolute | 1.277 ± 0.114 | 1.281 ± 0.039 | 1.365 ± 0.031 | 1.249 ± 0.037 | 1.422 ± 0.045 | 1.021 ± 0.058* |
| Relative | 3.50 ± 0.27 | 3.32 ± 0.09 | 3.42 ± 0.06 | 3.41 ± 0.12 | 3.77 ± 0.09 | 4.13 ± 0.12** |
| Liver | | | | | | |
| Absolute | 12.181 ± 0.531 | 13.017 ± 0.199 | 13.759 ± 0.382 | 12.599 ± 0.333 | 14.261 ± 0.349** | 11.782 ± 0.757 |
| Relative | 33.24 ± 0.46 | 33.76 ± 0.36 | 34.40 ± 0.66 | 34.16 ± 0.55 | 37.92 ± 0.71** | 47.23 ± 1.23** |
| Lung | | | | | | |
| Absolute | 1.570 ± 0.059 | 1.726 ± 0.053 | 1.745 ± 0.046 | 1.648 ± 0.036 | 1.638 ± 0.042 | 1.147 ± 0.045** |
| Relative | 4.30 ± 0.09 | 4.47 ± 0.10 | 4.37 ± 0.09 | 4.48 ± 0.09 | 4.36 ± 0.10 | 4.69 ± 0.15 |
| Prostate Gland | | | | | | |
| Absolute | 0.768 ± 0.096 | 0.812 ± 0.105 | 0.579 ± 0.043 | 0.799 ± 0.084 | 0.731 ± 0.091 | 0.504 ± 0.099 |
| Relative | 2.05 ± 0.21 | 2.10 ± 0.27 | 1.44 ± 0.09 | 2.19 ± 0.24 | 1.93 ± 0.23 | 1.96 ± 0.37 |
| Seminal Vesicle | | | | | | |
| Absolute | 1.387 ± 0.105 | 1.654 ± 0.103 | 1.798 ± 0.048* | 1.489 ± 0.114 | 1.538 ± 0.089 | 0.979 ± 0.104* |
| Relative | 3.78 ± 0.25 | 4.31 ± 0.29 | 4.51 ± 0.13 | 4.00 ± 0.26 | 4.11 ± 0.25 | 3.85 ± 0.29 |
| R. Testis | | | | | | |
| Absolute | 1.517 ± 0.043 | 1.581 ± 0.016 | 1.619 ± 0.029 | 1.559 ± 0.027 | 1.594 ± 0.024 | 0.457 ± 0.026** |
| Relative | 4.18 ± 0.12 | 4.11 ± 0.06 | 4.06 ± 0.08 | 4.24 ± 0.09 | 4.25 ± 0.08 | 1.86 ± 0.09** |
| Thymus | | | | | | |
| Absolute | 0.251 ± 0.023 | 0.263 ± 0.013 | 0.381 ± 0.115 | 0.259 ± 0.015 | 0.281 ± 0.017 | 0.182 ± 0.015 |
| Relative | 0.67 ± 0.04 | 0.68 ± 0.03 | 0.98 ± 0.32 | 0.71 ± 0.04 | 0.75 ± 0.05 | 0.74 ± 0.05 |

* 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).

TABLE D3
**Organ Weights and Organ-Weight-to-Body-Weight Ratios for Rats at the 15-Month Interim Evaluation in the 2-Year Feed Study of Butyl Benzyl Phthalate[a]**

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Male** | | | | |
| n | 10 | 10 | 10 | 10 |
| Necropsy body wt | 458 ± 10 | 460 ± 7 | 430 ± 9* | 416 ± 8** |
| R. Epididymis | | | | |
| Absolute | 0.428 ± 0.016 | 0.457 ± 0.023 | 0.473 ± 0.021 | 0.433 ± 0.009 |
| Relative | 0.93 ± 0.02 | 0.99 ± 0.05 | 1.10 ± 0.05* | 1.04 ± 0.02* |
| R. Kidney | | | | |
| Absolute | 1.608 ± 0.045 | 1.767 ± 0.054 | 1.667 ± 0.038 | 1.706 ± 0.048 |
| Relative | 3.52 ± 0.10 | 3.83 ± 0.08* | 3.88 ± 0.07** | 4.10 ± 0.08** |
| Liver | | | | |
| Absolute | 15.463 ± 0.524 | 16.120 ± 0.482 | 15.479 ± 0.382 | 15.761 ± 0.483 |
| Relative | 33.75 ± 0.92 | 35.00 ± 0.83 | 36.00 ± 0.55 | 37.82 ± 0.78** |
| R. Testis | | | | |
| Absolute | 1.638 ± 0.122 | 1.820 ± 0.180 | 1.580 ± 0.106 | 1.771 ± 0.165 |
| Relative | 3.62 ± 0.36 | 3.97 ± 0.41 | 3.69 ± 0.27 | 4.27 ± 0.42 |
| | **0 ppm** | **6,000 ppm** | **12,000 ppm** | **24,000 ppm** |
| **Female** | | | | |
| n | 10 | 10 | 10 | 10 |
| Necropsy body wt | 279 ± 7 | 279 ± 6 | 282 ± 5 | 215 ± 4** |
| R. Kidney | | | | |
| Absolute | 0.931 ± 0.022 | 1.004 ± 0.025 | 1.013 ± 0.022* | 0.868 ± 0.019 |
| Relative | 3.34 ± 0.07 | 3.61 ± 0.12 | 3.59 ± 0.05 | 4.04 ± 0.12** |
| Liver | | | | |
| Absolute | 8.629 ± 0.189 | 8.785 ± 0.230 | 9.301 ± 0.305 | 8.377 ± 0.153 |
| Relative | 31.01 ± 0.81 | 31.58 ± 0.90 | 33.00 ± 1.01 | 38.96 ± 0.57** |

* 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 AND HORMONE ASSAY RESULTS


TABLE E1 Hematology Data for Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate . . . . . 160

TABLE E2 Hematology Data for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate . . . . . 161

TABLE E3 Hematology and Hormone Assay Data for Rats at 6, 8, and 15 Months and at Study Termination in the 2-Year Feed Study of Butyl Benzyl Phthalate . . . . . 163

TABLE E1
**Hematology Data for Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate[a]**

| | 0 ppm | 300 ppm | 2,800 ppm | 25,000 ppm |
|---|---|---|---|---|
| n | 14 | 15 | 15 | 15 |
| Hematocrit (%) | 39.9 ± 0.5 | 39.8 ± 0.6 | 40.7 ± 0.5 | 38.2 ± 0.6 |
| Hemoglobin (g/dL) | 16.1 ± 0.2 | 16.0 ± 0.1 | 16.3 ± 0.2 | 15.5 ± 0.2 |
| Erythrocytes ($10^6/\mu L$) | 8.20 ± 0.09 | 8.22 ± 0.10 | 8.41 ± 0.09 | 7.25 ± 0.11** |
| Reticulocytes ($10^6/\mu L$) | 0.12 ± 0.01[b] | 0.12 ± 0.02[c] | 0.15 ± 0.01[c] | 0.18 ± 0.02[d] |
| Mean cell volume (fL) | 48.7 ± 0.1 | 48.4 ± 0.1 | 48.3 ± 0.1 | 52.7 ± 0.2** |
| Mean cell hemoglobin (pg) | 19.7 ± 0.1 | 19.5 ± 0.1 | 19.4 ± 0.2 | 21.5 ± 0.2** |
| Mean cell hemoglobin concentration (g/dL) | 40.4 ± 0.3 | 40.3 ± 0.3 | 40.1 ± 0.4 | 40.7 ± 0.3 |
| Platelets ($10^3/\mu L$) | 755.1 ± 10.8 | 738.5 ± 15.2 | 786.3 ± 14.5 | 963.5 ± 24.2** |
| Leukocytes ($10^3/\mu L$) | 7.36 ± 0.23 | 7.50 ± 0.26[e] | 7.58 ± 0.23 | 6.51 ± 0.21 |
| Segmented neutrophils ($10^3/\mu L$) | 1.33 ± 0.12[b] | 1.14 ± 0.08[f] | 1.31 ± 0.11[g] | 1.12 ± 0.05[d] |
| Lymphocytes ($10^3/\mu L$) | 6.29 ± 0.22[b] | 6.83 ± 0.19[f] | 6.47 ± 0.29[g] | 5.38 ± 0.24[d] |
| Monocytes ($10^3/\mu L$) | 0.08 ± 0.02[b] | 0.01 ± 0.01*[f] | 0.04 ± 0.03[g] | 0.05 ± 0.02[d] |
| Eosinophils ($10^3/\mu L$) | 0.04 ± 0.02[b] | 0.04 ± 0.04[f] | 0.05 ± 0.02[g] | 0.04 ± 0.02[d] |

* 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
[c] n=11
[d] n=12
[e] n=14
[f] n=7
[g] n=10

TABLE E2
Hematology Data for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 300 ppm | 900 ppm | 2,800 ppm | 8,300 ppm | 25,000 ppm |
|---|---|---|---|---|---|---|
| n | 13 | 14 | 15 | 14 | 15 | 11 |
| Hematocrit (%) | | | | | | |
| Day 30 | 43.3 ± 1.0[b] | 42.7 ± 0.8[b] | 45.0 ± 0.7 | 43.8 ± 1.1[b] | 42.3 ± 0.8 | 40.0 ± 1.1[b] |
| Day 60 | 37.9 ± 0.7[c] | 38.1 ± 0.6 | 38.1 ± 0.5 | 37.9 ± 0.3[b] | 36.8 ± 0.4 | 36.4 ± 0.5*[c] |
| Day 90 | 35.5 ± 0.6 | 37.1 ± 1.2 | 35.6 ± 1.1 | 36.2 ± 0.8 | 34.6 ± 0.8 | 31.7 ± 1.3* |
| Day 120 | 39.8 ± 1.1 | 40.9 ± 0.4 | 40.7 ± 0.7 | 40.2 ± 0.4 | 39.0 ± 0.5 | —[d] |
| Day 150 | 39.5 ± 1.1 | 40.6 ± 0.7 | 39.7 ± 0.5[c] | 38.3 ± 0.9 | 38.5 ± 0.6[e] | — |
| Day 180 | 37.8 ± 0.9[f] | 38.6 ± 0.9 | 39.5 ± 0.8[c] | 38.4 ± 1.0 | 37.6 ± 0.9[c] | 34.5 ± 0.7 |
| Hemoglobin (g/dL) | | | | | | |
| Day 30 | 16.2 ± 0.4[b] | 15.7 ± 0.3[b] | 16.8 ± 0.3 | 16.2 ± 0.3[b] | 16.2 ± 0.3 | 15.9 ± 0.4[b] |
| Day 60 | 15.3 ± 0.2[c] | 15.5 ± 0.2 | 15.5 ± 0.2 | 15.6 ± 0.1[b] | 15.3 ± 0.1 | 15.6 ± 0.2[c] |
| Day 90 | 14.2 ± 0.2 | 14.6 ± 0.4 | 14.2 ± 0.5 | 14.4 ± 0.2 | 14.1 ± 0.3 | 13.0 ± 0.6 |
| Day 120 | 15.5 ± 0.4 | 15.9 ± 0.2 | 15.9 ± 0.3 | 15.8 ± 0.2 | 15.5 ± 0.2 | — |
| Day 150 | 15.4 ± 0.4 | 15.8 ± 0.2 | 15.4 ± 0.2[c] | 15.0 ± 0.3 | 15.3 ± 0.2[c] | — |
| Day 180 | 14.5 ± 0.3[f] | 14.8 ± 0.3 | 15.1 ± 0.3[c] | 14.7 ± 0.4 | 14.7 ± 0.3 | 13.7 ± 0.3 |
| Erythrocytes ($10^6/\mu L$) | | | | | | |
| Day 30 | 8.02 ± 0.20[b] | 7.95 ± 0.16[b] | 8.33 ± 0.13 | 7.99 ± 0.18[b] | 7.83 ± 0.14 | 7.22 ± 0.20**[b] |
| Day 60 | 7.50 ± 0.11[c] | 7.61 ± 0.14 | 7.57 ± 0.09 | 7.54 ± 0.07[b] | 7.23 ± 0.05* | 6.71 ± 0.09**[c] |
| Day 90 | 7.20 ± 0.14 | 7.39 ± 0.20 | 7.15 ± 0.24 | 7.22 ± 0.09 | 6.89 ± 0.17 | 6.07 ± 0.27** |
| Day 120 | 8.05 ± 0.21 | 8.28 ± 0.08 | 8.17 ± 0.12 | 8.13 ± 0.09 | 7.81 ± 0.09* | — |
| Day 150 | 7.97 ± 0.22 | 8.20 ± 0.13 | 8.02 ± 0.09[c] | 7.74 ± 0.17* | 7.74 ± 0.11[c] | — |
| Day 180 | 7.71 ± 0.17[f] | 7.90 ± 0.19 | 8.06 ± 0.16[c] | 7.81 ± 0.22 | 7.67 ± 0.19[c] | 6.69 ± 0.13** |
| Reticulocytes ($10^6/\mu L$) | | | | | | |
| Day 30 | 0.09 ± 0.01[b] | 0.10 ± 0.01[b] | 0.06 ± 0.01 | 0.09 ± 0.01[b] | 0.11 ± 0.01 | 0.10 ± 0.01[b] |
| Day 60 | 0.12 ± 0.02[c] | 0.17 ± 0.03 | 0.13 ± 0.02 | 0.15 ± 0.01[b] | 0.13 ± 0.02 | 0.20 ± 0.02*[c] |
| Day 90 | 0.15 ± 0.01 | 0.20 ± 0.02 | 0.17 ± 0.02 | 0.14 ± 0.01 | 0.14 ± 0.02 | 0.26 ± 0.03* |
| Day 120 | 0.10 ± 0.02 | 0.10 ± 0.01 | 0.09 ± 0.01[c] | 0.10 ± 0.02[e] | 0.11 ± 0.01 | — |
| Day 150 | 0.15 ± 0.02 | 0.15 ± 0.01 | 0.13 ± 0.02[c] | 0.15 ± 0.01 | 0.14 ± 0.01[c] | — |
| Day 180 | 0.15 ± 0.01[f] | 0.15 ± 0.01 | 0.14 ± 0.02[c] | 0.12 ± 0.02 | 0.14 ± 0.02[c] | 0.17 ± 0.03 |
| Mean cell volume (fL) | | | | | | |
| Day 30 | 54.0 ± 0.3[b] | 53.7 ± 0.2[b] | 53.9 ± 0.3 | 54.7 ± 0.3[b] | 53.9 ± 0.2 | 55.3 ± 0.3**[b] |
| Day 60 | 50.5 ± 0.5[c] | 50.1 ± 0.4 | 50.3 ± 0.4 | 50.2 ± 0.2[b] | 50.9 ± 0.5 | 54.2 ± 0.3**[c] |
| Day 90 | 49.3 ± 0.3 | 50.1 ± 0.5 | 49.8 ± 0.4 | 50.1 ± 0.6 | 50.2 ± 0.2* | 52.4 ± 0.3** |
| Day 120 | 49.4 ± 0.2 | 49.4 ± 0.2 | 49.7 ± 0.3 | 49.5 ± 0.2 | 49.9 ± 0.2 | — |
| Day 150 | 49.6 ± 0.2 | 49.5 ± 0.3 | 49.5 ± 0.2[c] | 49.6 ± 0.2 | 49.7 ± 0.1[e] | — |
| Day 180 | 49.0 ± 0.4[f] | 48.8 ± 0.2 | 49.0 ± 0.2[c] | 49.2 ± 0.2 | 49.0 ± 0.1 | 51.6 ± 0.1** |
| Mean cell hemoglobin (pg) | | | | | | |
| Day 30 | 20.3 ± 0.1[b] | 19.8 ± 0.1[b] | 20.2 ± 0.1 | 20.3 ± 0.1[b] | 20.7 ± 0.2 | 22.0 ± 0.2**[b] |
| Day 60 | 20.5 ± 0.1[c] | 20.3 ± 0.2 | 20.5 ± 0.1 | 20.7 ± 0.1[b] | 21.1 ± 0.2** | 23.3 ± 0.2**[c] |
| Day 90 | 19.8 ± 0.1 | 19.8 ± 0.1 | 20.0 ± 0.2 | 20.0 ± 0.1 | 20.4 ± 0.1** | 21.4 ± 0.1** |
| Day 120 | 19.2 ± 0.1 | 19.2 ± 0.0 | 19.5 ± 0.1* | 19.4 ± 0.1* | 19.9 ± 0.1** | — |
| Day 150 | 19.4 ± 0.1 | 19.3 ± 0.1 | 19.3 ± 0.1[c] | 19.5 ± 0.1 | 19.8 ± 0.1**[c] | — |
| Day 180 | 18.8 ± 0.1[f] | 18.7 ± 0.1 | 18.7 ± 0.1[c] | 18.8 ± 0.1 | 19.1 ± 0.1*[c] | 20.5 ± 0.2** |
| Mean cell hemoglobin concentration (g/dL) | | | | | | |
| Day 30 | 37.5 ± 0.3[b] | 36.9 ± 0.3[b] | 37.4 ± 0.3 | 37.2 ± 0.3[b] | 38.4 ± 0.4 | 39.8 ± 0.3**[b] |
| Day 60 | 40.5 ± 0.3[c] | 40.6 ± 0.2 | 40.6 ± 0.2 | 41.3 ± 0.2*[b] | 41.5 ± 0.3* | 43.0 ± 0.4**[c] |
| Day 90 | 40.1 ± 0.2 | 39.5 ± 0.5 | 40.1 ± 0.2 | 40.0 ± 0.6 | 40.7 ± 0.1 | 40.9 ± 0.2** |
| Day 120 | 38.9 ± 0.1 | 38.8 ± 0.1 | 39.2 ± 0.1 | 39.2 ± 0.2 | 39.9 ± 0.2** | — |
| Day 150 | 39.1 ± 0.2 | 39.1 ± 0.2 | 38.9 ± 0.2[c] | 39.3 ± 0.2 | 39.6 ± 0.1*[e] | — |
| Day 180 | 38.4 ± 0.2[f] | 38.4 ± 0.2 | 38.2 ± 0.2[c] | 38.4 ± 0.2 | 39.0 ± 0.2*[c] | 39.8 ± 0.3** |

TABLE E2
**Hematology Data for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 300 ppm | 900 ppm | 2,800 ppm | 8,300 ppm | 25,000 ppm |
|---|---|---|---|---|---|---|
| n | 13 | 14 | 15 | 14 | 15 | 11 |
| Platelets ($10^3/\mu L$) | | | | | | |
| Day 30 | 914.9 ± 27.4[b] | 892.3 ± 17.3[b] | 881.8 ± 33.5 | 926.9 ± 26.5[b] | 954.6 ± 19.7 | 869.9 ± 16.2[b] |
| Day 60 | 743.1 ± 26.9[c] | 748.8 ± 20.1 | 735.5 ± 22.7 | 757.3 ± 16.4[b] | 731.8 ± 9.5 | 714.9 ± 30.0[c] |
| Day 90 | 767.5 ± 12.6 | 740.9 ± 18.4 | 751.7 ± 21.9 | 707.8 ± 11.1* | 743.1 ± 11.8[c] | 734.6 ± 28.6 |
| Day 120 | 676.4 ± 17.4 | 718.1 ± 21.3 | 713.9 ± 21.4 | 719.7 ± 15.0 | 709.7 ± 23.8 | — |
| Day 150 | 694.2 ± 25.9 | 679.2 ± 21.8 | 699.6 ± 16.5[c] | 695.9 ± 14.8 | 704.9 ± 16.5 | — |
| Day 180 | 636.8 ± 15.5[f] | 659.4 ± 13.0 | 656.6 ± 13.5[c] | 650.7 ± 14.2 | 676.1 ± 9.7 | 685.0 ± 12.7 |
| Leukocytes ($10^3/\mu L$) | | | | | | |
| Day 30 | 7.67 ± 0.39[b] | 7.74 ± 0.36[b] | 7.78 ± 0.40 | 7.98 ± 0.36[b] | 7.88 ± 0.34[c] | 8.96 ± 0.34[b] |
| Day 60 | 9.24 ± 0.25[c] | 8.42 ± 0.17 | 9.16 ± 0.31 | 8.57 ± 0.32[b] | 8.43 ± 0.26 | 9.57 ± 0.37[c] |
| Day 90 | 8.24 ± 0.38 | 8.34 ± 0.25 | 8.60 ± 0.33 | 7.81 ± 0.25 | 8.07 ± 0.18 | 8.44 ± 0.36 |
| Day 120 | 8.12 ± 0.36 | 8.72 ± 0.23 | 8.33 ± 0.27 | 8.12 ± 0.22 | 8.28 ± 0.29 | — |
| Day 150 | 9.39 ± 0.41[f] | 9.07 ± 0.31 | 9.30 ± 0.52[c] | 8.84 ± 0.23 | 9.37 ± 0.27 | — |
| Day 180 | 8.58 ± 0.19[f] | 8.88 ± 0.21 | 8.83 ± 0.42[c] | 9.45 ± 0.48 | 9.25 ± 0.33 | 9.25 ± 0.28 |
| Segmented neutrophils ($10^3/\mu L$) | | | | | | |
| Day 30 | 0.94 ± 0.13[b] | 0.89 ± 0.09[b] | 0.91 ± 0.08 | 1.00 ± 0.09[b] | 0.92 ± 0.10[c] | 0.87 ± 0.11[b] |
| Day 60 | 1.60 ± 0.17[c] | 1.33 ± 0.12 | 1.40 ± 0.11 | 1.41 ± 0.08[b] | 1.19 ± 0.08 | 0.99 ± 0.10**[c] |
| Day 90 | 1.50 ± 0.12 | 1.50 ± 0.13 | 1.48 ± 0.16 | 1.36 ± 0.18 | 1.58 ± 0.13 | 1.34 ± 0.08 |
| Day 120 | 1.47 ± 0.13 | 1.75 ± 0.12 | 1.37 ± 0.10 | 1.49 ± 0.15 | 1.58 ± 0.11 | — |
| Day 150 | 1.75 ± 0.16[f] | 1.40 ± 0.12 | 1.71 ± 0.11[c] | 1.60 ± 0.14 | 1.82 ± 0.17 | — |
| Day 180 | 1.53 ± 0.19[f] | 1.80 ± 0.11 | 1.47 ± 0.12[c] | 2.04 ± 0.18 | 2.01 ± 0.12 | 1.50 ± 0.09 |
| Lymphocytes ($10^3/\mu L$) | | | | | | |
| Day 30 | 6.60 ± 0.40[b] | 6.72 ± 0.34[b] | 6.72 ± 0.38 | 6.83 ± 0.37[b] | 6.83 ± 0.29[c] | 7.96 ± 0.29**[b] |
| Day 60 | 7.32 ± 0.16[c] | 6.86 ± 0.15 | 7.50 ± 0.26 | 6.99 ± 0.30[b] | 7.07 ± 0.23 | 8.39 ± 0.29[c] |
| Day 90 | 6.39 ± 0.32 | 6.52 ± 0.20 | 6.78 ± 0.28 | 6.21 ± 0.25 | 6.20 ± 0.12 | 6.77 ± 0.36 |
| Day 120 | 6.51 ± 0.35 | 6.76 ± 0.13 | 6.76 ± 0.25 | 6.47 ± 0.19 | 6.57 ± 0.27 | — |
| Day 150 | 7.45 ± 0.32[f] | 7.51 ± 0.25 | 7.38 ± 0.49[c] | 6.95 ± 0.23 | 7.43 ± 0.25 | — |
| Day 180 | 6.84 ± 0.24[f] | 6.90 ± 0.19 | 7.19 ± 0.36[c] | 7.23 ± 0.40 | 7.06 ± 0.32 | 7.47 ± 0.25 |
| Monocytes ($10^3/\mu L$) | | | | | | |
| Day 30 | 0.09 ± 0.02[b] | 0.07 ± 0.02[b] | 0.07 ± 0.01 | 0.07 ± 0.02[b] | 0.07 ± 0.02[c] | 0.09 ± 0.02[b] |
| Day 60 | 0.11 ± 0.03[c] | 0.06 ± 0.01 | 0.12 ± 0.03 | 0.08 ± 0.02[b] | 0.09 ± 0.03 | 0.10 ± 0.03[c] |
| Day 90 | 0.17 ± 0.04 | 0.12 ± 0.02 | 0.15 ± 0.03 | 0.10 ± 0.02 | 0.10 ± 0.02 | 0.16 ± 0.03 |
| Day 120 | 0.02 ± 0.01 | 0.06 ± 0.03 | 0.04 ± 0.01 | 0.04 ± 0.02 | 0.05 ± 0.02 | — |
| Day 150 | 0.09 ± 0.02[f] | 0.04 ± 0.02 | 0.04 ± 0.02[c] | 0.11 ± 0.02 | 0.02 ± 0.01 | — |
| Day 180 | 0.08 ± 0.03[f] | 0.06 ± 0.02 | 0.04 ± 0.01[c] | 0.06 ± 0.02 | 0.05 ± 0.02 | 0.15 ± 0.07 |
| Eosinophils ($10^3/\mu L$) | | | | | | |
| Day 30 | 0.03 ± 0.01[b] | 0.06 ± 0.01[b] | 0.07 ± 0.02 | 0.06 ± 0.02[b] | 0.07 ± 0.03[c] | 0.04 ± 0.01[b] |
| Day 60 | 0.10 ± 0.02[c] | 0.10 ± 0.03 | 0.07 ± 0.02 | 0.05 ± 0.02*[b] | 0.05 ± 0.02* | 0.05 ± 0.02[c] |
| Day 90 | 0.14 ± 0.03 | 0.11 ± 0.03 | 0.12 ± 0.02 | 0.12 ± 0.03 | 0.12 ± 0.04 | 0.12 ± 0.03 |
| Day 120 | 0.10 ± 0.03 | 0.11 ± 0.03 | 0.15 ± 0.03 | 0.09 ± 0.02 | 0.09 ± 0.02 | — |
| Day 150 | 0.08 ± 0.03[f] | 0.11 ± 0.03 | 0.14 ± 0.03[c] | 0.16 ± 0.04 | 0.09 ± 0.02 | — |
| Day 180 | 0.11 ± 0.03[f] | 0.11 ± 0.03 | 0.09 ± 0.02[c] | 0.10 ± 0.02 | 0.12 ± 0.04 | 0.11 ± 0.04 |

* Significantly different ($P \le 0.05$) from the control group by Dunn's or Shirley's test
** $P \le 0.01$
[a] Mean ± standard error. Statistical tests were performed on unrounded data.
[b] n=15
[c] n=14
[d] Not measured at this time point
[e] n=13
[f] n=12

TABLE E3
Hematology and Hormone Assay Data for Rats at 6, 8, and 15 Months and at Study Termination in the 2-Year Feed Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Male** | | | | |
| Hematology | | | | |
| 6 Months | | | | |
| n | 10 | 10 | 10 | 10 |
| Hematocrit (%) | 47.1 ± 0.4 | 47.7 ± 0.5 | 46.7 ± 0.5 | 46.7 ± 0.4 |
| Hemoglobin (g/dL) | 15.2 ± 0.1 | 15.3 ± 0.1 | 15.0 ± 0.1 | 15.1 ± 0.1 |
| Erythrocytes ($10^6/\mu L$) | 9.28 ± 0.08 | 9.31 ± 0.07 | 9.13 ± 0.08 | 9.08 ± 0.06* |
| Reticulocytes ($10^6/\mu L$) | 0.11 ± 0.01 | 0.09 ± 0.01 | 0.11 ± 0.02 | 0.09 ± 0.01 |
| Nucleated erythrocytes ($10^3/\mu L$) | 0.08 ± 0.03 | 0.03 ± 0.02 | 0.07 ± 0.03 | 0.06 ± 0.02 |
| Mean cell volume (fL) | 50.8 ± 0.3 | 51.3 ± 0.3 | 51.2 ± 0.2 | 51.5 ± 0.4 |
| Mean cell hemoglobin (pg) | 16.4 ± 0.1 | 16.4 ± 0.1 | 16.5 ± 0.1 | 16.7 ± 0.0** |
| Mean cell hemoglobin concentration (g/dL) | 32.3 ± 0.2 | 32.1 ± 0.1 | 32.1 ± 0.1 | 32.5 ± 0.2 |
| Platelets ($10^3/\mu L$) | 594.8 ± 12.5 | 586.3 ± 10.9 | 595.9 ± 22.2 | 616.2 ± 21.6 |
| Leukocytes ($10^3/\mu L$) | 10.58 ± 0.38 | 9.99 ± 0.35 | 10.38 ± 0.57 | 10.63 ± 0.55 |
| Segmented neutrophils ($10^3/\mu L$) | 1.85 ± 0.24 | 1.77 ± 0.15 | 2.04 ± 0.19 | 2.34 ± 0.40 |
| Lymphocytes ($10^3/\mu L$) | 8.15 ± 0.24 | 7.92 ± 0.36 | 7.80 ± 0.48 | 7.74 ± 0.29 |
| Atypical lymphocytes ($10^3/\mu L$) | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 |
| Monocytes ($10^3/\mu L$) | 0.32 ± 0.06 | 0.22 ± 0.06 | 0.44 ± 0.10 | 0.42 ± 0.07 |
| Eosinophils ($10^3/\mu L$) | 0.26 ± 0.05 | 0.09 ± 0.04* | 0.09 ± 0.02 | 0.12 ± 0.04 |
| 15 Months | | | | |
| n | 9 | 10 | 10 | 10 |
| Hematocrit (%) | 42.8 ± 0.7 | 45.1 ± 0.8 | 44.0 ± 0.4 | 43.7 ± 0.6 |
| Hemoglobin (g/dL) | 14.3 ± 0.2 | 15.0 ± 0.3 | 14.6 ± 0.1 | 14.6 ± 0.1 |
| Erythrocytes ($10^6/\mu L$) | 8.40 ± 0.13 | 8.75 ± 0.15 | 8.58 ± 0.08 | 8.45 ± 0.09 |
| Reticulocytes ($10^6/\mu L$) | 0.13 ± 0.02 | 0.12 ± 0.01 | 0.13 ± 0.01 | 0.15 ± 0.02 |
| Nucleated erythrocytes ($10^3/\mu L$) | 0.20 ± 0.10 | 0.04 ± 0.03 | 0.04 ± 0.02 | 0.03 ± 0.02 |
| Mean cell volume (fL) | 51.0 ± 0.4 | 51.5 ± 0.3 | 51.3 ± 0.3 | 51.6 ± 0.4 |
| Mean cell hemoglobin (pg) | 17.0 ± 0.1 | 17.2 ± 0.1 | 17.1 ± 0.1 | 17.2 ± 0.1 |
| Mean cell hemoglobin concentration (g/dL) | 33.3 ± 0.3 | 33.3 ± 0.2 | 33.2 ± 0.1 | 33.4 ± 0.2 |
| Platelets ($10^3/\mu L$) | 731.6 ± 36.9 | 715.4 ± 18.2 | 723.4 ± 24.9 | 734.4 ± 14.5 |
| Leukocytes ($10^3/\mu L$) | 8.29 ± 0.55 | 8.59 ± 0.74 | 8.05 ± 0.59 | 8.43 ± 0.63 |
| Segmented neutrophils ($10^3/\mu L$) | 3.26 ± 0.39 | 3.23 ± 0.27 | 3.47 ± 0.37 | 3.42 ± 0.31 |
| Lymphocytes ($10^3/\mu L$) | 4.64 ± 0.33 | 4.80 ± 0.49 | 4.12 ± 0.36 | 4.60 ± 0.46 |
| Atypical lymphocytes ($10^3/\mu L$) | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 |
| Monocytes ($10^3/\mu L$) | 0.27 ± 0.03 | 0.41 ± 0.09 | 0.32 ± 0.07 | 0.34 ± 0.06 |
| Eosinophils ($10^3/\mu L$) | 0.11 ± 0.04 | 0.14 ± 0.05 | 0.13 ± 0.05 | 0.08 ± 0.03 |

**TABLE E3**
**Hematology and Hormone Assay Data for Rats at 6, 8, and 15 Months and at Study Termination in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 3,000 ppm | 6,000 ppm | 12,000 ppm |
|---|---|---|---|---|
| **Male** (continued) | | | | |
| Hormone Assays | | | | |
| n | 10 | 10 | 10 | 10 |
| Thyroid-stimulating hormone (ng/mL) | | | | |
| 6 Months | 3 ± 1[b] | 8 ± 6 | 6 ± 2[c] | 4 ± 1 |
| 8 Months | 1 ± 0 | 3 ± 0** | 2 ± 0 | 2 ± 0 |
| 15 Months | 1 ± 0 | 1 ± 0 | 2 ± 0* | 1 ± 0 |
| Study termination | 2 ± 0[b] | 1 ± 0 | 1 ± 0 | 2 ± 0 |
| Triiodothyronine (ng/dL) | | | | |
| 6 Months | 80 ± 9[c] | 100 ± 7[b] | 82 ± 7[d] | 77 ± 12[d] |
| 15 Months | 115 ± 7[c] | 114 ± 4[b] | 123 ± 4[b] | 116 ± 2[c] |
| Study termination | 117 ± 10 | 80 ± 11 | 121 ± 17[c] | 117 ± 11[c] |
| Thyroxine (μg/dL) | | | | |
| 6 Months | 4 ± 0[e] | 4 ± 0[c] | 4 ± 0[c] | 4 ± 0[c] |
| 15 Months | 4 ± 1 | 4 ± 0 | 5 ± 0 | 3 ± 0[b] |
| Study termination | 4 ± 0 | 3 ± 0 | 4 ± 0 | 4 ± 0 |
| Adrenocorticotropic hormone (pg/mL) | | | | |
| 6 Months | 3,102 ± 542 | 3,420 ± 636 | 3,014 ± 462 | 1,450 ± 365* |
| 15 Months | 1,263 ± 310[b] | 1,599 ± 452[b] | 2,299 ± 494 | 2,754 ± 757 |
| Study termination | 741 ± 122 | 641 ± 91 | 704 ± 130 | 835 ± 122 |
| Testosterone (ng/mL) | | | | |
| 6 Months | 1 ± 0[f] | 1 ± 0[g] | 1[h] | 2 ± 0*[f] |
| 8 Months | 1 ± 0 | 1 ± 0 | 2 ± 0 | 2 ± 0[b] |
| 15 Months | 2 ± 0[d] | 2 ± 1[i] | 2 ± 1[b] | 2 ± 0[e] |
| Study termination | 5 ± 5 | 1 ± 0 | 1 ± 0 | 1 ± 0* |

TABLE E3
Hematology and Hormone Assay Data for Rats at 6, 8, and 15 Months and at Study Termination in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Female** | | | | |
| Hematology | | | | |
| 6 Months | | | | |
| n | 9 | 10 | 10 | 10 |
| Hematocrit (%) | 45.4 ± 0.4 | 46.0 ± 0.4 | 43.7 ± 0.9 | 45.1 ± 0.5 |
| Hemoglobin (g/dL) | 14.9 ± 0.1 | 15.1 ± 0.1 | 14.5 ± 0.3 | 15.0 ± 0.2 |
| Erythrocytes ($10^6/\mu L$) | 8.31 ± 0.08 | 8.42 ± 0.07 | 8.11 ± 0.18 | 8.32 ± 0.10 |
| Reticulocytes ($10^6/\mu L$) | 0.12 ± 0.02 | 0.09 ± 0.01 | 0.13 ± 0.02 | 0.15 ± 0.02 |
| Nucleated erythrocytes ($10^3/\mu L$) | 0.07 ± 0.02 | 0.07 ± 0.03 | 0.07 ± 0.02 | 0.04 ± 0.02 |
| Mean cell volume (fL) | 54.6 ± 0.2 | 54.6 ± 0.3 | 53.9 ± 0.3 | 54.2 ± 0.3 |
| Mean cell hemoglobin (pg) | 17.9 ± 0.1 | 17.9 ± 0.1 | 17.9 ± 0.1 | 18.1 ± 0.1 |
| Mean cell hemoglobin concentration (g/dL) | 32.8 ± 0.1 | 32.8 ± 0.2 | 33.2 ± 0.2 | 33.3 ± 0.1 |
| Platelets ($10^3/\mu L$) | 645.8 ± 45.0 | 608.2 ± 17.5 | 573.0 ± 14.7 | 566.1 ± 17.7 |
| Leukocytes ($10^3/\mu L$) | 7.46 ± 0.82 | 7.59 ± 0.58 | 7.43 ± 0.37 | 8.17 ± 0.58 |
| Segmented neutrophils ($10^3/\mu L$) | 1.58 ± 0.45 | 1.35 ± 0.18 | 1.50 ± 0.14 | 1.04 ± 0.12 |
| Lymphocytes ($10^3/\mu L$) | 5.41 ± 0.35 | 5.85 ± 0.38 | 5.55 ± 0.35 | 6.62 ± 0.46 |
| Atypical lymphocytes ($10^3/\mu L$) | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 |
| Monocytes ($10^3/\mu L$) | 0.33 ± 0.07 | 0.30 ± 0.07 | 0.30 ± 0.05 | 0.41 ± 0.06 |
| Eosinophils ($10^3/\mu L$) | 0.15 ± 0.06 | 0.09 ± 0.02 | 0.08 ± 0.02 | 0.09 ± 0.03 |
| 15 Months | | | | |
| n | 9 | 10 | 10 | 10 |
| Hematocrit (%) | 44.9 ± 0.6 | 45.3 ± 0.4 | 44.7 ± 0.8 | 42.8 ± 0.6* |
| Hemoglobin (g/dL) | 15.1 ± 0.3 | 15.5 ± 0.1 | 15.2 ± 0.2 | 14.7 ± 0.2 |
| Erythrocytes ($10^6/\mu L$) | 7.91 ± 0.15 | 8.12 ± 0.08 | 7.96 ± 0.15 | 7.79 ± 0.11 |
| Reticulocytes ($10^6/\mu L$) | 0.15 ± 0.02 | 0.15 ± 0.02 | 0.15 ± 0.01 | 0.13 ± 0.02 |
| Nucleated erythrocytes ($10^3/\mu L$) | 0.28 ± 0.06 | 0.16 ± 0.04 | 0.13 ± 0.02 | 0.47 ± 0.10 |
| Mean cell volume (fL) | 56.8 ± 0.5 | 55.8 ± 0.3 | 56.2 ± 0.2 | 55.0 ± 0.2** |
| Mean cell hemoglobin (pg) | 19.1 ± 0.1 | 19.1 ± 0.1 | 19.1 ± 0.1 | 18.8 ± 0.1 |
| Mean cell hemoglobin concentration (g/dL) | 33.7 ± 0.2 | 34.2 ± 0.1 | 33.9 ± 0.1 | 34.3 ± 0.1* |
| Platelets ($10^3/\mu L$) | 628.9 ± 50.8 | 696.5 ± 24.0 | 664.4 ± 23.7 | 691.9 ± 21.5 |
| Leukocytes ($10^3/\mu L$) | 4.20 ± 0.26 | 5.20 ± 0.69 | 4.97 ± 0.26 | 6.34 ± 0.66** |
| Segmented neutrophils ($10^3/\mu L$) | 1.36 ± 0.19 | 1.57 ± 0.28 | 1.22 ± 0.11 | 1.28 ± 0.15 |
| Lymphocytes ($10^3/\mu L$) | 2.75 ± 0.15 | 3.48 ± 0.61 | 3.62 ± 0.23* | 4.87 ± 0.59** |
| Atypical lymphocytes ($10^3/\mu L$) | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.02 ± 0.02 |
| Monocytes ($10^3/\mu L$) | 0.04 ± 0.01 | 0.08 ± 0.03 | 0.06 ± 0.02 | 0.12 ± 0.02 |
| Eosinophils ($10^3/\mu L$) | 0.05 ± 0.01 | 0.08 ± 0.02 | 0.07 ± 0.02 | 0.06 ± 0.02 |

**TABLE E3**
**Hematology and Hormone Assay Data for Rats at 6, 8, and 15 Months and at Study Termination in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| | 0 ppm | 6,000 ppm | 12,000 ppm | 24,000 ppm |
|---|---|---|---|---|
| **Female** (continued) | | | | |
| Hormone Assays | | | | |
| n | 10 | 10 | 10 | 10 |
| Thyroid-stimulating hormone (ng/mL) | | | | |
| 6 Months | 2 ± 1 | 1 ± 0[b] | 2 ± 2[b] | 0 ± 0 |
| 15 Months | 1 ± 0 | 1 ± 0 | 2 ± 0 | 2 ± 0 |
| Study termination | 1 ± 0 | 1 ± 0[b] | 1 ± 0 | 1 ± 0 |
| Triiodothyronine (ng/dL) | | | | |
| 6 Months | 86 ± 8[i] | 99 ± 7[e] | 82 ± 5[c] | 60 ± 4* |
| 15 Months | 126 ± 5 | 125 ± 3 | 117 ± 4 | 98 ± 5** |
| Study termination | 130 ± 20[e] | 101 ± 6[g] | 97 ± 6[g] | 83 ± 5*[d] |
| Thyroxine (μg/dL) | | | | |
| 6 Months | 4 ± 0 | 4 ± 0[b] | 4 ± 0[b] | 3 ± 0 |
| 15 Months | 3 ± 0 | 3 ± 0 | 3 ± 0 | 2 ± 0** |
| Study termination | 3 ± 0 | 3 ± 0 | 3 ± 0 | 3 ± 0 |
| Adrenocorticotropic hormone (pg/mL) | | | | |
| 6 Months | 1,946 ± 328 | 1,912 ± 413 | 1,838 ± 255 | 1,966 ± 311 |
| 15 Months | 1,565 ± 368 | 816 ± 88 | 1,609 ± 303 | 731 ± 106[b] |
| Study termination | 666 ± 76 | 496 ± 68 | 634 ± 78 | 334 ± 35** |
| Prolactin (ng/mL) | | | | |
| 15 Months | 26 ± 3 | 42 ± 11 | 37 ± 11 | 28 ± 6 |
| Study termination | 86 ± 15[f] | 131 ± 25[e] | 172 ± 37[g] | 80 ± 42[e] |

* 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
[c] n=8
[d] n=5
[e] n=6
[f] n=2
[g] n=4
[h] n=1; no standard error calculated
[i] n=7

# APPENDIX F
# REPRODUCTIVE TISSUE EVALUATIONS AND MODIFIED MATING STUDY RESULTS


TABLE F1 Summary of Reproductive Tissue Evaluations for Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate . . . . . 168

TABLE F2 Fertility, Maternal Body Weight, and Developmental Toxicity Data for Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate . . . . . 169

TABLE F3 Summary of Reproductive Tissue Evaluations for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate . . . . . 170

TABLE F1
**Summary of Reproductive Tissue Evaluations for Male Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate[a]**

| | 0 ppm | 300 ppm | 2,800 ppm | 25,000 ppm |
|---|---|---|---|---|
| n | 15 | 14 | 15 | 15 |
| Weights (g) | | | | |
| Necropsy body wt | 317 ± 4 | 315 ± 4[b] | 310 ± 4 | 225 ± 5** |
| R. cauda | 0.221 ± 0.040 | 0.196 ± 0.023 | 0.179 ± 0.005 | 0.068 ± 0.005**[c] |
| R. epididymis | 0.541 ± 0.042 | 0.505 ± 0.026[b] | 0.486 ± 0.013 | 0.230 ± 0.009** |
| R. testis | 1.497 ± 0.017 | 1.378 ± 0.081[b] | 1.502 ± 0.019 | 0.442 ± 0.014** |
| Epididymal spermatozoal parameters | | | | |
| Motility (%) | 79.21 ± 3.38 | 73.24 ± 6.14[b] | 76.49 ± 3.29 | —[d] |
| Concentration ($10^6$/g cauda epididymal tissue) | 373.94 ± 39.52 | 324.14 ± 49.56 | 261.47 ± 39.50* | 0.567 ± 0.39** |
| Abnormal (%) | 0.987 ± 0.106 | 0.857 ± 0.064 | 0.960 ± 0.105 | — |

* Significantly different ($P \leq 0.05$) from the control group by Williams' or Dunnett's test (organ weights and body weights) or by Dunn's or Shirley's test (epididymal spermatozoal parameters)
** $P \leq 0.01$
[a] Data are presented as mean ± standard error.
[b] n=15
[c] n=14
[d] Not measured at this exposure level due to there being too few sperm to assess

TABLE F2
Fertility, Maternal Body Weight, and Developmental Toxicity Data for Rats in the 10-Week Modified Mating Study of Butyl Benzyl Phthalate[a]

| | 0 ppm | 300 ppm | 2,800 ppm | 25,000 ppm |
|---|---|---|---|---|
| Fertility | | | | |
| Male fertility index[b] | 23/30 (77%) | 25/30 (83%) | 28/30 (93%) | 10/30 (33%)** |
| Female fertility index[c] | 20/23 (87%) | 23/25 (92%) | 23/28 (82%) | 0/0 (0%)** |
| Days of mating[d] | 4.47 ± 0.38 | 4.30 ± 0.41 | 4.83 ± 0.28 | 6.23 ± 0.21** |
| Maternal body wt (g) | | | | |
| n | 19 | 21 | 23 | 30[e] |
| Mating day 0 | 191 ± 3 | 195 ± 2 | 188 ± 2 | 192 ± 2 |
| Gestation day 0 | 200 ± 3 | 202 ± 3 | 198 ± 2 | |
| Gestation day 6 | 208 ± 3 | 211 ± 2 | 207 ± 2 | |
| Gestation day 9 | 215 ± 3 | 218 ± 3 | 213 ± 2 | |
| Gestation day 13 | 225 ± 3 | 228 ± 3 | 224 ± 2 | |
| Developmental Toxicity | | | | |
| n | 20 | 23 | 23 | |
| Litters with dead fetuses | 0 (0%) | 2 (9%) | 1 (4%) | |
| Live fetuses/total fetuses | 203/203 | 189/192 | 232/233 | |
| Live fetuses/litter | 10.15 ± 0.83 | 8.22 ± 0.94 | 10.09 ± 0.60 | |
| Corpora lutea/litter | 12.65 ± 0.36 | 12.13 ± 0.59 | 12.22 ± 0.38 | |
| Implantations/litter | 10.60 ± 0.83 | 8.78 ± 0.92 | 10.83 ± 0.64 | |
| Litters with resorptions | 6 (30%) | 7 (30%) | 13 (57%) | |
| Resorptions/litter | 0.50 ± 0.22 | 0.44 ± 0.15 | 0.70 ± 0.16 | |

** Significantly different ($P \leq 0.01$) from the control group by the Chi-square test or Dunn's or Shirley's test

[a] Data for days of mating, maternal body weights, and live fetuses, corpora lutea, implantations, and resorptions per litter are presented as mean ± standard error. Maternal body weight differences from the control group are not significant by Dunnett's or Williams' test. Differences from the control group for developmental toxicity data are not significant by the Chi-square test or Dunn's or Shirley's test.

[b] Females determined to be sperm positive at the end of the mating period/females housed with males

[c] Females pregnant at necropsy/females initially determined to be sperm positive

[d] Days until females were determined to be sperm positive (measured for 30 females per exposure group)

[e] No litters at this exposure level

TABLE F3
**Summary of Reproductive Tissue Evaluations for Male Rats in the 26-Week Feed Study of Butyl Benzyl Phthalate[a]**

| | 0 ppm | 300 ppm | 8,300 ppm | 25,000 ppm |
|---|---|---|---|---|
| n | 12 | 14 | 15 | 11 |
| Weights (g) | | | | |
| Necropsy body wt | 367 ± 16 | 386 ± 5 | 369 ± 8[b] | 249 ± 13** |
| R. cauda | 0.227 ± 0.020 | 0.197 ± 0.007 | 0.212 ± 0.007 | 0.109 ± 0.009** |
| R. epididymis | 0.531 ± 0.019 | 0.549 ± 0.018 | 0.559 ± 0.014 | 0.282 ± 0.025** |
| R. testis | 1.517 ± 0.043 | 1.581 ± 0.016 | 1.559 ± 0.027[b] | 0.457 ± 0.026** |
| Epididymal spermatozoal parameters | | | | |
| Motility (%) | 74.15 ± 9.62 | 75.49 ± 8.42 | 75.94 ± 6.28 | —[c] |
| Concentration ($10^6$/g cauda epididymal tissue) | 284.52 ± 32.23 | 285.75 ± 40.18 | 376.79 ± 22.68 | 2.06 ± 2.06** |
| Abnormal (%) | 1.017 ± 0.097 | 0.879 ± 0.115 | 0.840 ± 0.094 | — |

** Significantly different ($P \leq 0.01$) from the control group by Williams' or Dunnett's test (organ weights and body weights) or by Dunn's or Shirley's test (epididymal spermatozoal parameters)
[a] Data are presented as mean ± standard error.
[b] n=14
[c] Not measured at this exposure level due to there being too few sperm to assess

# APPENDIX G
# CHEMICAL CHARACTERIZATION AND DOSE FORMULATION STUDIES


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# CHEMICAL CHARACTERIZATION AND DOSE FORMULATION STUDIES

## PROCUREMENT AND CHARACTERIZATION OF BUTYL BENZYL PHTHALATE

Butyl benzyl phthalate was obtained from Chem Central (Kansas City, MO) in two lots (C090882 and L-121 1-87). Lot C090882 was used during the 10-week modified mating study and the 26-week study, and lot L-121 1-87 was used during the 2-year study. 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 butyl benzyl phthalate studies are on file at the National Institute of Environmental Health Sciences.

Both lots of the chemical, a clear, colorless, viscous liquid, were identified as butyl benzyl phthalate by infrared, ultraviolet/visible, and nuclear magnetic resonance spectroscopy. All spectra were consistent with the structure of butyl benzyl phthalate, and the infrared and nuclear magnetic resonance spectra were consistent with the literature spectra (*Sadtler Standard Spectra*) of butyl benzyl phthalate (Figures G1 and G2).

The purity of both lots was determined by elemental analyses, Karl Fischer water analysis, functional group titrations (free acid titration and ester hydrolysis), thin-layer chromatography (TLC), and gas chromatography. For one type of functional group titration, samples of butyl benzyl phthalate were diluted with methanol and titrated with 0.01 N sodium hydroxide. The titration was monitored potentiometrically with a combination mV/pH electrode filled with aqueous 3 M potassium chloride. For the second type of functional group titration, samples of butyl benzyl phthalate were dissolved in excess aqueous 1 N potassium hydroxide and 2-propanol. After gentle agitation, the excess potassium hydroxide was neutralized with 0.5 N hydrochloric acid. The titration was monitored visually with phenolphthalein indicator or potentiometrically with a combination pH/mV electrode filled with aqueous 3 M potassium chloride. TLC was performed on Silica Gel 60 F-254 plates with two solvent systems: 1) *n*-hexane: *p*-dioxane (90:10) and 2) methylene chloride. Plates were examined under visible light and/or short wavelength (254 nm) ultraviolet light and with a spray of 20% antimony (V) chloride in carbon tetrachloride or chloroform. For lot C090882, the plates were heated at 110° C for 20 minutes. Dimethyl terephthalate was used as a reference standard. Gas chromatography was performed using a flame ionization detector. For each lot, two systems were used:

A) 3% SP-2100 on 100/120 Supelcoport glass column, with a nitrogen carrier gas at a flow rate of 70 mL/minute and an oven temperature program of 50° C for 5 minutes, then 50° to 270° C at 10° C per minute, and

B) 3% SP-2401 on 100/120 Supelcoport glass column, with a nitrogen carrier gas at a flow rate of 70 mL/minute and an oven temperature program of 50° C for 5 minutes, then 50° to 250° C at 10° C per minute.

For lot L-121 1-87, an additional system was also used:

C) DB-5 Megabore fused silica column, with a helium carrier gas at a flow rate of 10 mL/minute, a nitrogen make-up gas at a flow rate of 20 mL/minute, and an oven temperature program of 150° to 300° C at 10° C per minute with a 17-minute final hold.

For lot C090882, elemental analyses for carbon and hydrogen were in agreement with the theoretical values for butyl benzyl phthalate. Karl Fischer water analysis indicated 0.23% ± 0.01% water. Functional group

titrations indicated free acid values of 0.011% ± 0.001% and 0.00158 ± 0.0004 mEq acid/g sample and purities of 99.5% ± 0.2% and 100.2% ± 0.4%. TLC indicated a major spot and a slight trace impurity by system 1 and only a major spot by system 2. Gas chromatography using system A indicated one major peak and four impurities with a cumulative area of 3.0% relative to the major peak. The largest impurity had an area estimated to be 2.3% relative to the major peak. Gas chromatography using system B indicated one major peak and four impurities with a cumulative area of 2.8% relative to the major peak. Another analysis of lot C090882 by gas chromatography using system B indicated one major peak and three impurities with a cumulative area of 2.27% relative to the major peak. The largest impurity had an area estimated to be 1.76% relative to the major peak. The largest impurity was identified as benzyl phthalate by gas chromatography using system A, and the sample contained 1.43% ± 0.03% benzyl phthalate. Major peak comparisons of lot C090882 with lot M2676 with system A but with an isothermal oven temperature of 250° C indicated a purity of 100.3% ± 0.8% for lot C090882 relative to lot M2676. The overall purity of lot C090882 was determined to be greater than 97%.

For lot L-121 1-87, elemental analyses for carbon and hydrogen were in agreement with the theoretical values for butyl benzyl phthalate. Karl Fischer water analysis indicated 0.11% ± 0.01% water. Functional group titrations indicated 0.00112 ± 0.00003 mEq of acid/g of sample and a purity of 99.1 ± 0.4%. TLC indicated a major spot and two slight trace impurities by system 1 and a major spot and a trace impurity by system 2. Gas chromatography using system B with a final oven temperature of 270° C with a 20-minute final hold indicated one major peak and five impurities with a combined area of 1.20% relative to the major peak. Gas chromatography using system C indicated one major peak and three impurities with a combined area of 1.04% relative to the major peak. Major peak comparisons of lot L-121 1-87 with lot M2676 with system A but with an isothermal oven temperature of 250° C and *n*-pentacosane as an internal standard indicated a purity of 99.3% ± 0.4% for lot L-121 1-87 relative to lot M2676. The overall purity of lot L-121 1-87 was determined to be greater than or equal to 99%.

Stability studies of the bulk chemical were performed by the analytical chemistry laboratory. Gas chromatography was performed using system A except with an isothermal oven temperature of 250° C and *n*-pentacosane as an internal standard. These studies indicated that butyl benzyl phthalate 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 the dark at 4° C in tightly sealed 1-gallon amber-glass bottles during the 10-week modified mating study and the 26-week study and in the dark at approximately 22° C in 1-gallon amber-glass bottles with Teflon®-lined lids during the 2-year study. Stability was monitored using gas chromatography during the 10-week modified mating study, the 26-week study, and the 2-year study. No significant degradation of the bulk chemical was detected.

## Preparation and Analysis of Dose Formulations

The dose formulations were prepared weekly by mixing butyl benzyl phthalate with feed (Table G1). A butyl benzyl phthalate/feed premix was prepared by hand and then blended with feed in a Patterson-Kelly twin-shell blender for 25 minutes using an intensifier bar for the initial 15 minutes for the 10-week modified mating study and for the 26-week study and for 15 minutes using an intensifier bar for the initial 5 minutes for the 2-year study. Formulations were stored in plastic bags inside containers at 5° C for up to 4 weeks.

Homogeneity studies of the 300, 6,250, and 25,000 ppm dose formulations were performed by the study laboratory and the analytical chemistry laboratory. In addition, homogeneity studies of the 3,000 and 24,000 ppm dose formulations were performed by the study laboratory. The samples were taken in triplicate from the three blender ports. For the 300, 3,000, 24,000 and 25,000 ppm formulations, the samples were extracted with hexane and clarified by centrifugation. Aliquots of the extracts were shaken with 0.1 N sodium hydroxide and were centrifuged to separate the two phases. For each sample, an aliquot

from the hexane layer was pipetted into a vial and evaporated under a stream of nitrogen. An internal standard solution of octaphenone in acetonitrile:water (80:20) was added, and the solutions were purified and then analyzed using high-performance liquid chromatography (HPLC). HPLC was performed with a Brownlee MPLC RP-18 column using ultraviolet detection (280 nm) and a mobile phase of water:acetonitrile (15:85, 25:75, 30:70, or 40:60) at a flow rate of 1.0 or 1.4 mL/minute. For the 6,250 ppm formulation, the samples were extracted with acetonitrile and clarified by centrifugation. Aliquots of the extracts were mixed with water and an internal standard solution of octaphenone in acetonitrile and were further diluted with acetonitrile. The solutions were filtered and analyzed using the same HPLC system used for the 300, 3,000, 24,000 and 25,000 ppm formulations but with a mobile phase ratio of 30:70 and a flow rate of 1.4 mL/minute. Stability studies of the 300 and 6,250 ppm dose formulations were also performed using the same HPLC methods described for the homogeneity analyses of the 300 and 6,250 ppm formulations, respectively. Homogeneity was confirmed and the stability of the dose formulations was confirmed for at least 4 weeks at temperatures up to 5° C when stored protected from light.

Periodic analyses of the dose formulations of butyl benzyl phthalate were conducted at the study laboratories and analytical chemistry laboratory using HPLC. For the 10-week modified mating study, the 26-week study, and the 2-year study, the formulations were analyzed every 8 to 9 weeks (Tables G2 and G3). All of the dose formulations analyzed were within 10% of the target concentration. Results of periodic referee analyses performed by the analytical chemistry laboratory agreed with the results obtained by the study laboratory (Table G4).

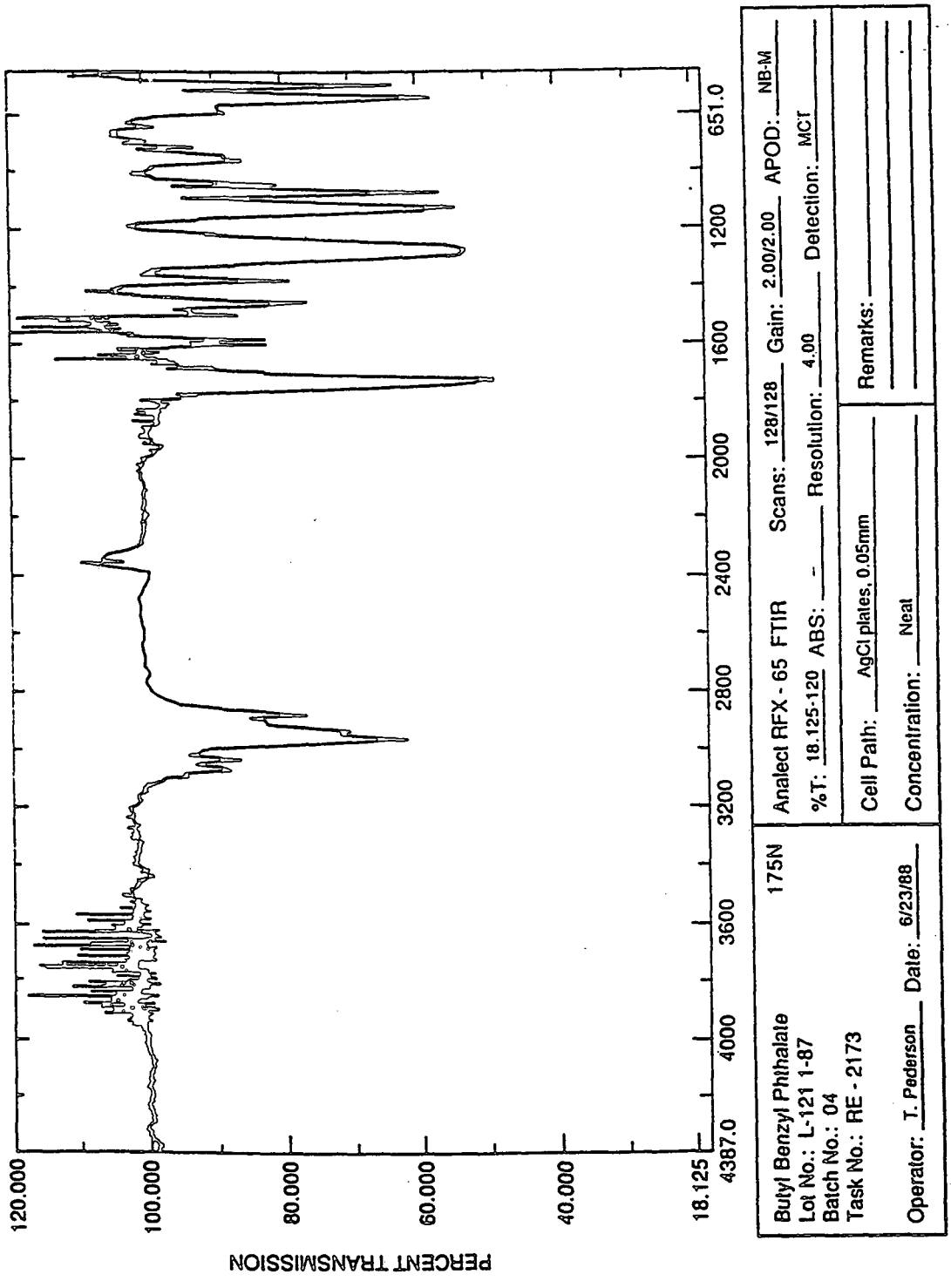


FIGURE G1
Infrared Absorption Spectrum of Butyl Benzyl Phthalate

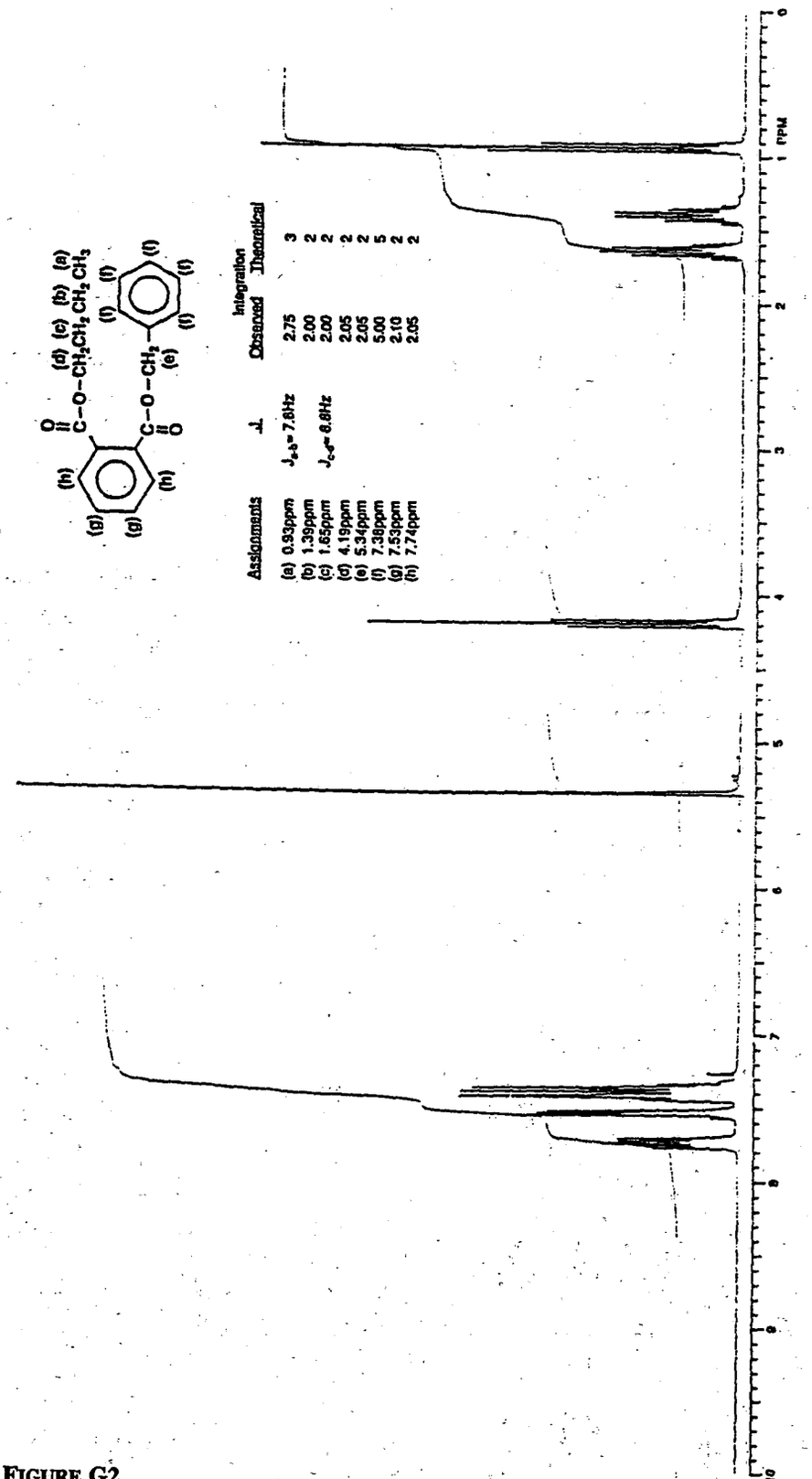


**FIGURE G2**
**Nuclear Magnetic Resonance Spectrum of Butyl Benzyl Phthalate**

TABLE G1
Preparation and Storage of Dose Formulations in the Feed Studies of Butyl Benzyl Phthalate

| 10-Week Modified Mating Study | 26-Week Study | 2-Year Study |
|---|---|---|
| **Preparation** | | |
| A premix of feed and butyl benzyl phthalate was prepared, then layered into the remaining feed and blended in a Patterson-Kelly twin-shell blender with the intensifier bar on for 15 minutes and off for 10 minutes. Doses were prepared weekly. | A premix of feed and butyl benzyl phthalate was prepared, then layered into the remaining feed and blended in a Patterson-Kelly twin-shell blender with the intensifier bar on for 15 minutes and off for 10 minutes. Doses were prepared weekly. | A premix of feed and butyl benzyl phthalate 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 weekly. |
| **Chemical Lot Number** | | |
| C090882 | C090882 | L-121 1-87 |
| **Maximum Storage Time** | | |
| 4 weeks | 4 weeks | 4 weeks |
| **Storage Conditions** | | |
| Stored in plastic bags placed in rigid containers at 5° C | Stored in plastic bags placed in rigid containers at 5° C | Stored in double-thickness plastic bags placed in rigid plastic containers at -20° C or 5° C |
| **Study Laboratory** | | |
| Hazleton Laboratories America, Inc. (Rockville, MD) | Hazleton Laboratories America, Inc. (Rockville, MD) | Southern Research Institute (Birmingham, AL) |
| **Referee Laboratory** | | |
| Midwest Research Institute (Kansas City, MO) | Midwest Research Institute (Kansas City, MO) | Midwest Research Institute (Kansas City, MO) |

TABLE G2
**Results of Analysis of Dose Formulations Administered to Rats in the 10-Week Modified Mating Study and in the 26-Week Feed Study of Butyl Benzyl Phthalate**

| Date Prepared | Date Analyzed | Target Concentration (ppm) | Determined Concentration[a] (ppm) | Difference from Target (%) |
|---|---|---|---|---|
| 19 September 1984 | 22 September 1984 | 300 | 310[b] | +3 |
| | | 300 | 310[c] | +3 |
| | | 300 | 310[d] | +3 |
| | | 300 | 308[b] | +3 |
| | | 300 | 308[c] | +3 |
| | | 300 | 310[d] | +3 |
| | | 900 | 850 | -6 |
| | | 900 | 852 | -5 |
| | | 2,800 | 2,790 | 0 |
| | | 2,800 | 2,790 | 0 |
| | | 8,300 | 7,680 | -7 |
| | | 25,000 | 24,400[b] | -2 |
| | | 25,000 | 24,400[c] | -2 |
| | | 25,000 | 24,400[d] | -2 |
| 13 November 1984 | 14 November 1984 | 300 | 321 | +7 |
| | | 900 | 973 | +8 |
| | | 2,800 | 2,800 | 0 |
| | | 8,300 | 8,450 | +2 |
| | | 25,000 | 23,900 | -4 |
| 8 January 1985 | 9 January 1985 | 300 | 285 | -5 |
| | | 900 | 850 | -6 |
| | | 2,800 | 2,690 | -4 |
| | | 8,300 | 7,830 | -6 |
| | | 25,000 | 23,600 | -6 |
| 12 March 1985 | 13 March 1985 | 300 | 299 | 0 |
| | | 900 | 891 | -1 |
| | | 2,800 | 2,630 | -6 |
| | | 8,300 | 7,830 | -6 |
| | | 25,000 | 23,900 | -4 |

[a] Results of duplicate analyses
[b] Sample selection from top left of twin-shell blender
[c] Sample selection from top right of twin-shell blender
[d] Sample selection from bottom of twin-shell blender

TABLE G3
Results of Analysis of Dose Formulations Administered to Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate

| Date Prepared | Date Analyzed | Target Concentration (ppm) | Determined Concentration[a] (ppm) | Difference from Target (%) |
|---|---|---|---|---|
| 19 June 1989 | 20 June 1989 | 3,000 | 2,860 | -5 |
| | | 6,000 | 5,700 | -5 |
| | | 6,000 | 5,500 | -8 |
| | | 12,000 | 11,400 | -5 |
| | | 12,000 | 11,600 | -3 |
| | | 24,000 | 23,200 | -3 |
| 21 August 1989 | 22-23 August 1989 | 3,000 | 3,000 | 0 |
| | | 6,000 | 5,960 | -1 |
| | | 6,000 | 5,980 | 0 |
| | | 12,000 | 12,300 | +3 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 12,200 | +2 |
| | | 24,000 | 24,100 | 0 |
| | | 24,000 | 24,800 | +3 |
| 23 October 1989 | 24-25 October 1989 | 3,000 | 3,020 | +1 |
| | | 6,000 | 5,960 | -1 |
| | | 6,000 | 5,980 | 0 |
| | | 12,000 | 12,200 | +2 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 12,200 | +2 |
| | | 24,000 | 24,100 | 0 |
| | | 24,000 | 23,700 | -1 |
| 18 December 1989 | 18-19 December 1989 | 3,000 | 2,940 | -2 |
| | | 6,000 | 6,040 | +1 |
| | | 6,000 | 6,000 | 0 |
| | | 12,000 | 11,900 | -1 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 12,000 | 0 |
| | | 24,000 | 24,600 | +3 |
| | | 24,000 | 24,200 | +1 |
| 12 February 1990 | 13 February 1990 | 3,000 | 2,960 | -1 |
| | | 6,000 | 5,940 | -1 |
| | | 6,000 | 5,940 | -1 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 11,900 | -1 |
| | | 24,000 | 24,300 | +1 |
| | | 24,000 | 24,200 | +1 |
| 26 March 1990 | 27-28 March 1990 | 3,000 | 2,820 | -6 |
| | | 6,000 | 5,920 | -1 |
| | | 6,000 | 5,870 | -2 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 11,600 | -3 |
| | | 12,000 | 12,300 | +3 |
| | | 24,000 | 23,800 | -1 |
| | | 24,000 | 23,000 | -4 |

TABLE G3
**Results of Analysis of Dose Formulations Administered to Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate** (continued)

| Date Prepared | Date Analyzed | Target Concentration (ppm) | Determined Concentration (ppm) | Difference from Target (%) |
|---|---|---|---|---|
| 21 May 1990 | 21-22 May 1990 | 3,000 | 2,870 | -4 |
| | | 6,000 | 5,960 | -1 |
| | | 6,000 | 5,990 | 0 |
| | | 12,000 | 11,740 | -2 |
| | | 12,000 | 11,990 | 0 |
| | | 12,000 | 12,170 | +1 |
| | | 24,000 | 23,910 | 0 |
| | | 24,000 | 23,630 | -2 |
| 23 July 1990 | 24-27 July 1990 | 3,000 | 3,110 | +4 |
| | | 6,000 | 6,540 | +9 |
| | | 6,000 | 6,370 | +6 |
| | | 12,000 | 12,450 | +4 |
| | | 12,000 | 12,620 | +5 |
| | | 12,000 | 12,590 | +5 |
| | | 24,000 | 24,370 | +2 |
| | | 24,000 | 24,510 | +2 |
| 24 September 1990 | 25 September 1990-2 October 1990 | 3,000 | 2,870 | -4 |
| | | 6,000 | 5,910 | -2 |
| | | 6,000 | 5,890 | -2 |
| | | 12,000 | 11,800 | -2 |
| | | 12,000 | 11,800 | -2 |
| | | 12,000 | 11,700 | -3 |
| | | 24,000 | 23,400 | -3 |
| | | 24,000 | 23,400 | -3 |
| 26 November 1990 | 27 November 1990 | 3,000 | 2,970 | -1 |
| | | 6,000 | 5,980 | 0 |
| | | 6,000 | 6,020 | 0 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 12,000 | 0 |
| | | 24,000 | 23,800 | -1 |
| | | 24,000 | 23,900 | 0 |
| 21 January 1991 | 22 January 1991 | 3,000 | 3,120 | +4 |
| | | 6,000 | 5,600 | -7 |
| | | 6,000 | 5,770 | -4 |
| | | 12,000 | 12,000 | 0 |
| | | 12,000 | 11,630 | -3 |
| | | 12,000 | 12,080 | +1 |
| | | 24,000 | 23,920 | 0 |
| | | 24,000 | 23,820 | -1 |

TABLE G3
Results of Analysis of Dose Formulations Administered to Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate (continued)

| Date Prepared | Date Analyzed | Target Concentration (ppm) | Determined Concentration (ppm) | Difference from Target (%) |
|---|---|---|---|---|
| 18 March 1991 | 18-19 March 1991 | 3,000 | 2,990 | 0 |
| | | 6,000 | 5,900 | -2 |
| | | 6,000 | 5,730 | -5 |
| | | 6,000 | 5,950 | -1 |
| | | 12,000 | 11,600 | -3 |
| | | 12,000 | 11,800 | -2 |
| | | 12,000 | 11,900 | -1 |
| | | 24,000 | 23,700 | -1 |
| | | 24,000 | 23,600 | -2 |
| | | 24,000 | 23,500 | -2 |
| 13 May 1991 | 13-15 May 1991 | 3,000 | 3,020 | +1 |
| | | 6,000 | 5,910 | -2 |
| | | 6,000 | 5,930 | -1 |
| | | 6,000 | 5,970 | -1 |
| | | 12,000 | 11,900 | -1 |
| | | 12,000 | 11,900 | -1 |
| | | 12,000 | 12,000 | 0 |
| | | 24,000 | 24,100 | 0 |
| | | 24,000 | 24,000 | 0 |
| | | 24,000 | 24,000 | 0 |
| 8 July 1991 | 9-10 July 1991 | 6,000 | 5,980 | 0 |
| | | 12,000 | 12,000 | 0 |
| | | 24,000 | 24,100 | 0 |
| 9 September 1991 | 9-10 September 1991 | 6,000 | 5,820 | -3 |
| | | 12,000 | 12,000 | 0 |
| | | 24,000 | 23,800 | -1 |
| 4 November 1991 | 4-5 November 1991 | 6,000 | 6,110 | +2 |
| | | 12,000 | 11,500 | -4 |
| | | 24,000 | 24,200 | +1 |
| 6 January 1992 | 6 January 1992 | 6,000 | 5,970 | -1 |
| | | 12,000 | 11,700 | -3 |
| | | 24,000 | 23,600 | -2 |
| 9 March 1992 | 9-10 March 1992 | 6,000 | 5,940 | -1 |
| | | 12,000 | 11,900 | -1 |
| | | 24,000 | 24,000 | 0 |

[a] Results of duplicate analyses

TABLE G4
**Results of Referee Analysis of Dose Formulations Administered to Rats in the 10-Week Modified Mating Study and the 26-Week and 2-Year Feed Studies of Butyl Benzyl Phthalate**

| Date Prepared | Target Concentration (ppm) | Determined Concentration (ppm) | |
|---|---|---|---|
| | | Study Laboratory[a] | Referee Laboratory[b] |
| **10-Week Modified Mating Study and 26-Week Study (Hazleton Laboratories America, Inc.)** | | | |
| 19 September 1984 | 300 | 308 | 305 ± 3 |
| 8 January 1985 | 8,300 | 7,830 | 7,960 ± 120 |
| **2-Year Study (Southern Research Institute)** | | | |
| 19 June 1989 | 12,000 | 11,400 | 11,800 ± 500 |

[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 BUTYL BENZYL PHTHALATE

TABLE H1 Feed and Compound Consumption by Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate . . . . . 184
TABLE H2 Feed and Compound Consumption by Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate . . . . . 185

TABLE H1
**Feed and Compound Consumption by Male Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate**

| Week | 0 ppm | | 3,000 ppm | | | 6,000 ppm | | | 12,000 ppm | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | 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 | 14.6 | 149 | 15.1 | 148 | 308 | 14.8 | 147 | 606 | 14.1 | 141 | 1,198 |
| 3 | 15.9 | 183 | | | | | | | | | |
| 4 | 15.6 | 205 | | | | | | | | | |
| 5 | 17.8 | 233 | 19.3 | 236 | 245 | 17.8 | 233 | 456 | 19.7 | 226 | 1,043 |
| 6 | 17.5 | 253 | 17.9 | 255 | 210 | 17.1 | 253 | 404 | 18.4 | 243 | 905 |
| 7 | 16.5 | 266 | | | | | | | | | |
| 8 | 17.4 | 286 | | | | | | | | | |
| 9 | 16.5 | 298 | 15.6 | 299 | 156 | 15.2 | 292 | 313 | 15.6 | 284 | 660 |
| 10 | 16.2 | 310 | 16.8 | 310 | 163 | 17.0 | 301 | 339 | 16.6 | 291 | 686 |
| 11 | 15.3 | 319 | | | | | | | | | |
| 12 | 16.4 | 326 | | | | | | | | | |
| 13 | 16.4 | 339 | 16.6 | 338 | 147 | 17.2 | 321 | 321 | 16.8 | 315 | 642 |
| 17 | 17.3 | 370 | 16.3 | 367 | 134 | 12.4 | 350 | 212 | 12.3 | 338 | 438 |
| 21 | 15.9 | 391 | 17.8 | 389 | 137 | 18.6 | 367 | 305 | 17.5 | 351 | 599 |
| 25 | 20.5 | 391 | 16.4 | 403 | 122 | 17.0 | 381 | 268 | 15.5 | 362 | 512 |
| 29 | 14.7 | 416 | 15.6 | 411 | 114 | 15.1 | 394 | 231 | 14.4 | 377 | 458 |
| 33 | 14.7 | 423 | 16.2 | 421 | 115 | 14.3 | 399 | 214 | 15.6 | 382 | 490 |
| 37 | 15.0 | 429 | 15.6 | 429 | 109 | 16.1 | 410 | 236 | 15.2 | 389 | 469 |
| 41 | 17.1 | 437 | 16.4 | 434 | 114 | 15.8 | 422 | 225 | 16.5 | 394 | 502 |
| 45 | 17.5 | 443 | | | | 16.3 | 429 | 228 | 17.0 | 405 | 503 |
| 49 | 16.1 | 449 | 16.0 | 449 | 107 | 16.0 | 433 | 221 | 15.9 | 413 | 463 |
| 53 | 16.3 | 451 | 17.1 | 451 | 114 | 15.6 | 437 | 215 | 16.7 | 414 | 484 |
| 57 | 15.3 | 450 | 15.2 | 453 | 101 | 16.0 | 432 | 222 | 16.2 | 412 | 470 |
| 61 | 15.7 | 451 | 17.4 | 445 | 117 | 16.0 | 434 | 221 | 16.1 | 413 | 468 |
| 65 | 15.4 | 451 | 16.0 | 446 | 107 | 15.7 | 434 | 217 | 15.1 | 420 | 431 |
| 69 | 14.2 | 446 | 15.5 | 445 | 104 | 14.5 | 440 | 198 | 15.1 | 419 | 434 |
| 73 | 15.1 | 448 | 14.5 | 447 | 97 | 14.6 | 441 | 199 | 15.4 | 427 | 433 |
| 77 | 14.1 | 446 | 15.3 | 443 | 104 | 13.8 | 441 | 188 | 14.4 | 428 | 404 |
| 81 | 13.5 | 440 | 13.2 | 437 | 91 | 11.9 | 431 | 166 | 13.3 | 422 | 378 |
| 85 | 13.6 | 440 | 14.2 | 431 | 99 | 13.8 | 424 | 195 | 14.3 | 418 | 409 |
| 89 | 13.9 | 439 | 14.0 | 430 | 97 | 14.2 | 428 | 199 | 13.6 | 413 | 396 |
| 93 | | | | | | 13.1 | 422 | 186 | 13.0 | 422 | 370 |
| 97 | 14.5 | 433 | 13.5 | 428 | 95 | 12.4 | 410 | 182 | 12.7 | 411 | 372 |
| 101 | 12.4 | 426 | 12.8 | 419 | 92 | 13.9 | 407 | 204 | 11.6 | 402 | 347 |
| **Mean for weeks** | | | | | | | | | | | |
| 1-13 | 16.3 | 264 | 16.9 | 264 | 205 | 16.5 | 258 | 407 | 16.9 | 250 | 856 |
| 14-52 | 16.5 | 417 | 16.3 | 413 | 119 | 15.7 | 398 | 238 | 15.5 | 379 | 493 |
| 53-101 | 14.5 | 443 | 14.9 | 440 | 101 | 14.3 | 429 | 199 | 14.4 | 417 | 415 |

[a] Grams of feed consumed per animal per day
[b] Milligrams of butyl benzyl phthalate consumed per kilogram body weight per day

TABLE H2
Feed and Compound Consumption by Female Rats in the 2-Year Feed Study of Butyl Benzyl Phthalate

| Week | 0 ppm | | 6,000 ppm | | | 12,000 ppm | | | 24,000 ppm | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | 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 | 10.3 | 121 | 11:1 | 121 | 551 | 10.9 | 122 | 1,076 | 7.5 | 114 | 1,582 |
| 3 | 11.0 | 135 | | | | | | | | | |
| 4 | 11.3 | 142 | | | | | | | | | |
| 5 | 11.1 | 153 | 12.6 | 151 | 499 | 11.8 | 149 | 951 | | | |
| 6 | 10.6 | 159 | 11.6 | 162 | 431 | 11.0 | 157 | 838 | 11.3 | 151 | 1,791 |
| 7 | 10.9 | 167 | | | | | | | | | |
| 8 | 10.9 | 171 | | | | | | | | | |
| 9 | 11.3 | 175 | | | | | | | | | |
| 10 | 10.9 | 178 | 10.8 | 179 | 362 | 10.9 | 175 | 749 | 10.5 | 171 | 1,478 |
| 11 | 10.8 | 180 | | | | | | | | | |
| 12 | 11.0 | 186 | | | | | | | | | |
| 13 | 10.3 | 185 | 10.5 | 187 | 337 | 10.1 | 179 | 677 | 9.8 | 176 | 1,338 |
| 17 | 10.2 | 198 | 10.4 | 197 | 317 | 10.2 | 193 | 634 | 9.7 | 183 | 1,274 |
| 21 | 10.3 | 204 | 10.0 | 204 | 292 | 10.2 | 196 | 623 | 9.9 | 187 | 1,269 |
| 25 | | | 10.4 | 213 | 293 | 9.9 | 206 | 575 | 10.1 | 193 | 1,254 |
| 29 | 9.3 | 218 | 9.8 | 219 | 269 | 9.6 | 211 | 547 | 9.0 | 197 | 1,090 |
| 33 | 9.7 | 223 | 10.2 | 225 | 272 | 9.4 | 216 | 520 | 9.3 | 199 | 1,127 |
| 37 | 10.7 | 231 | 10.9 | 231 | 282 | 10.7 | 224 | 575 | 9.6 | 203 | 1,133 |
| 41 | 10.7 | 238 | | | | 11.2 | 230 | 581 | | | |
| 45 | 11.4 | 240 | 11.4 | 244 | 281 | 11.5 | 239 | 579 | 9.7 | 207 | 1,119 |
| 49 | 12.5 | 258 | 11.4 | 261 | 262 | 11.8 | 248 | 570 | 10.4 | 212 | 1,177 |
| 53 | 11.6 | 269 | 12.1 | 270 | 270 | 11.9 | 258 | 553 | 10.1 | 214 | 1,130 |
| 57 | 11.1 | 276 | 11.9 | 275 | 260 | 11.8 | 266 | 534 | 10.6 | 214 | 1,184 |
| 61 | 11.4 | 281 | 11.6 | 280 | 249 | 11.4 | 270 | 506 | 9.7 | 216 | 1,073 |
| 65 | 11.7 | 286 | 11.6 | 287 | 243 | 11.6 | 277 | 502 | 10.1 | 220 | 1,104 |
| 69 | 11.1 | 296 | 11.3 | 293 | 233 | 11.5 | 281 | 491 | 9.9 | 222 | 1,068 |
| 73 | 11.1 | 298 | 10.9 | 301 | 218 | 11.2 | 284 | 473 | 9.8 | 226 | 1,046 |
| 77 | 11.3 | 300 | 11.2 | 304 | 221 | 11.4 | 287 | 475 | 9.8 | 230 | 1,028 |
| 81 | 11.7 | 300 | 11.4 | 303 | 227 | 11.2 | 291 | 462 | 10.0 | 222 | 1,084 |
| 85 | 11.8 | 312 | 11.7 | 308 | 228 | 12.0 | 299 | 480 | 10.8 | 231 | 1,126 |
| 89 | 12.1 | 321 | 12.4 | 321 | 231 | 11.7 | 306 | 460 | 10.6 | 237 | 1,074 |
| 93 | 10.6 | 320 | 11.8 | 321 | 221 | 11.1 | 308 | 432 | 10.5 | 236 | 1,069 |
| 97 | 11.4 | 319 | 11.8 | 323 | 219 | 12.1 | 309 | 469 | 10.4 | 233 | 1,070 |
| 101 | 10.2 | 318 | 11.5 | 325 | 211 | 11.7 | 307 | 457 | 10.1 | 232 | 1,049 |
| **Mean for weeks** | | | | | | | | | | | |
| 1-13 | 10.9 | 163 | 11.3 | 160 | 436 | 10.9 | 156 | 858 | 9.8 | 153 | 1,547 |
| 14-52 | 10.6 | 226 | 10.6 | 224 | 284 | 10.5 | 218 | 578 | 9.7 | 198 | 1,180 |
| 53-101 | 11.3 | 300 | 11.6 | 301 | 233 | 11.6 | 288 | 484 | 10.2 | 226 | 1,085 |

[a] Grams of feed consumed per animal per day
[b] Milligrams of butyl benzyl phthalate 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 | 188 |
| TABLE I2 | Vitamins and Minerals in NIH-07 Rat and Mouse Ration | 188 |
| TABLE I3 | Nutrient Composition of NIH-07 Rat and Mouse Ration | 189 |
| TABLE I4 | Contaminant Levels in NIH-07 Rat and Mouse Ration | 190 |

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[b] | Range | Number of Samples |
|---|---|---|---|
| Protein (% by weight) | 23.60 ± 0.71 | 21.30 — 25.20 | 32 |
| Crude fat (% by weight) | 5.45 ± 0.32 | 4.80 — 5.90 | 32 |
| Crude fiber (% by weight) | 3.49 ± 0.31 | 3.00 — 4.10 | 32 |
| Ash (% by weight) | 6.82 ± 0.33 | 6.01 — 7.27 | 32 |
| **Amino Acids (% of total diet)** | | | |
| Arginine | 1.287 ± 0.084 | 1.100 — 1.390 | 10 |
| Cystine | 0.306 ± 0.075 | 0.181 — 0.400 | 10 |
| Glycine | 1.160 ± 0.050 | 1.060 — 1.220 | 10 |
| Histidine | 0.580 ± 0.024 | 0.531 — 0.608 | 10 |
| Isoleucine | 0.917 ± 0.034 | 0.867 — 0.965 | 10 |
| Leucine | 1.972 ± 0.052 | 1.850 — 2.040 | 10 |
| Lysine | 1.273 ± 0.051 | 1.200 — 1.370 | 10 |
| Methionine | 0.437 ± 0.115 | 0.306 — 0.699 | 10 |
| Phenylalanine | 0.994 ± 0.125 | 0.665 — 1.110 | 10 |
| Threonine | 0.896 ± 0.055 | 0.824 — 0.985 | 10 |
| Tryptophan | 0.223 ± 0.160 | 0.107 — 0.671 | 10 |
| Tyrosine | 0.677 ± 0.105 | 0.564 — 0.794 | 10 |
| Valine | 1.089 ± 0.057 | 0.962 — 1.170 | 10 |
| **Essential Fatty Acids (% of total diet)** | | | |
| Linoleic | 2.389 ± 0.233 | 1.830 — 2.570 | 9 |
| Linolenic | 0.277 ± 0.036 | 0.210 — 0.320 | 9 |
| **Vitamins** | | | |
| Vitamin A (IU/kg) | 6,966 ± 1,512 | 4,290 — 12,540 | 32 |
| Vitamin D (IU/kg) | 4,450 ± 1,382 | 3,000 — 6,300 | 4 |
| α-Tocopherol (ppm) | 36.92 ± 9.32 | 22.5 — 48.9 | 9 |
| Thiamine (ppm) | 18.94 ± 2.06 | 15.0 — 25.0 | 32 |
| Riboflavin (ppm) | 7.92 ± 0.93 | 6.10 — 9.00 | 10 |
| Niacin (ppm) | 100.95 ± 25.92 | 65.0 — 150.0 | 9 |
| Pantothenic acid (ppm) | 30.30 ± 3.60 | 23.0 — 34.6 | 10 |
| Pyridoxine (ppm) | 9.25 ± 2.62 | 5.60 — 14.0 | 10 |
| Folic acid (ppm) | 2.51 ± 0.64 | 1.80 — 3.70 | 10 |
| Biotin (ppm) | 0.267 ± 0.049 | 0.19 — 0.35 | 10 |
| Vitamin $B_{12}$ (ppb) | 40.14 ± 20.04 | 10.6 — 65.0 | 10 |
| Choline (ppm) | 3,068 ± 314 | 2,400 — 3,430 | 9 |
| **Minerals** | | | |
| Calcium (%) | 1.19 ± 0.09 | 1.02 — 1.37 | 32 |
| Phosphorus (%) | 0.94 ± 0.05 | 0.80 — 1.03 | 32 |
| Potassium (%) | 0.887 ± 0.067 | 0.772 — 0.971 | 8 |
| Chloride (%) | 0.526 ± 0.092 | 0.380 — 0.635 | 8 |
| Sodium (%) | 0.315 ± 0.034 | 0.258 — 0.370 | 10 |
| Magnesium (%) | 0.168 ± 0.008 | 0.151 — 0.180 | 10 |
| Sulfur (%) | 0.274 ± 0.063 | 0.208 — 0.420 | 10 |
| Iron (ppm) | 356.2 ± 90.0 | 255.0 — 523.0 | 10 |
| Manganese (ppm) | 92.24 ± 5.35 | 81.70 — 99.40 | 10 |
| Zinc (ppm) | 58.14 ± 9.91 | 46.10 — 81.60 | 10 |
| Copper (ppm) | 11.50 ± 2.40 | 8.090 — 15.39 | 10 |
| Iodine (ppm) | 3.70 ± 1.14 | 1.52 — 5.83 | 10 |
| Chromium (ppm) | 1.71 ± 0.45 | 0.85 — 2.09 | 9 |
| Cobalt (ppm) | 0.797 ± 0.23 | 0.490 — 1.150 | 6 |

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

| Nutrient | Mean ± Standard Deviation[b] | Range | Number of Samples |
|---|---|---|---|
| **Contaminants** | | | |
| Arsenic (ppm) | 0.40 ± 0.18 | 0.10 — 0.80 | 32 |
| Cadmium (ppm) | 0.11 ± 0.06 | 0.05 — 0.20 | 32 |
| Lead (ppm) | 0.33 ± 0.38 | 0.10 — 2.10 | 32 |
| Mercury (ppm) | 0.03 ± 0.01 | 0.02 — 0.05 | 32 |
| Selenium (ppm)[c] | 0.35 ± 0.10 | 0.20 — 0.55 | 31 |
| Aflatoxins (ppb)[d] | <5.0 | | 31 |
| Nitrate nitrogen (ppm)[e] | 11.60 ± 5.15 | 1.80 — 20.0 | 32 |
| Nitrite nitrogen (ppm)[e] | 0.24 ± 0.20 | 0.10 — 1.00 | 32 |
| BHA (ppm)[f] | 1.42 ± 0.85 | 1.00 — 4.0 | 31 |
| BHT (ppm)[f] | 1.29 ± 1.10 | 1.00 — 7.00 | 31 |
| Aerobic plate count (CFU/g)[f] | 102,209 ± 92,146 | 4,700 — 380,000 | 32 |
| Coliform (MPN/g)[g] | 20.50 ± 22.30 | 3.00 — 93.00 | 32 |
| *Escherichia coli* (MPN/g) | 3.25 ± 1.08 | 3.00 — 9.0 | 32 |
| Total nitrosoamines (ppb)[g] | 7.05 ± 2.60 | 2.00 — 13.70 | 32 |
| *N*-Nitrosodimethylamine (ppb)[g] | 5.12 ± 1.60 | 1.00 — 9.40 | 32 |
| *N*-Nitrosopyrrolidine (ppb)[g] | 1.68 ± 1.25 | 0.00 — 4.70 | 32 |
| **Pesticides (ppm)** | | | |
| α-BHC | <0.01 | | 31 |
| β-BHC | <0.02 | | 31 |
| γ-BHC | <0.01 | | 31 |
| δ-BHC | <0.01 | | 31 |
| Heptachlor | <0.01 | | 31 |
| Aldrin | <0.01 | | 31 |
| Heptachlor epoxide | <0.01 | | 31 |
| DDE | <0.01 | | 31 |
| DDD | <0.01 | | 31 |
| DDT | <0.01 | | 31 |
| HCB | <0.01 | | 31 |
| Mirex | <0.01 | | 31 |
| Methoxychlor | <0.05 | | 31 |
| Dieldrin | <0.01 | | 31 |
| Endrin | <0.01 | | 31 |
| Telodrin | <0.01 | | 31 |
| Chlordane | <0.05 | | 31 |
| Toxaphene | <0.1 | | 31 |
| Estimated PCBs | <0.2 | | 31 |
| Ronnel | <0.01 | | 31 |
| Ethion | <0.02 | | 31 |
| Trithion | <0.05 | | 31 |
| Diazinon | <0.1 | | 31 |
| Methyl parathion | <0.02 | | 31 |
| Ethyl parathion | <0.02 | | 31 |
| Malathion | 0.28 ± 0.26 | <0.05 — 1.00 | 34 |
| Endosulfan I | <0.01 | | 31 |
| Endosulfan II | <0.01 | | 31 |
| Endosulfan sulfate | <0.03 | | 31 |

[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 4 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

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# 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 10-week modified mating study, the 26-week study, and the 2-year study. 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 |
|---|---|
| **10-Week Modified Mating Study** | |
| ELISA | |
| *Mycoplasma arthritidis* | Study termination |
| *Mycoplasma pulmonis* | Study termination |
| PVM (pneumonia virus of mice) | Study termination |
| RCV/SDA (rat coronavirus/ sialodacryoadenitis virus) | Study termination |
| Sendai | Study termination |
| Hemagglutination Inhibition | |
| H-1 (Toolan's H-1 virus) | Study termination |
| KRV (Kilham rat virus) | Study termination |
| **26-Week Study** | |
| ELISA | |
| *M. arthritidis* | 13 weeks, study termination |
| *M. pulmonis* | 13 weeks, study termination |
| PVM | 13 weeks, study termination |
| RCV/SDA | 13 weeks, study termination |
| Sendai | 13 weeks, study termination |
| Hemagglutination Inhibition | |
| H-1 | 13 weeks, study termination |
| KRV | 13 weeks, study termination |

| Method and Test | Time of Analysis |
|---|---|
| **2-Year Study** | |
| ELISA | |
| *M. arthritidis* | Study termination |
| *M. pulmonis* | Study termination |
| PVM | 6, 12, 14, and 17 months, study termination |
| RCV/SDA | 6, 12, 14, and 17 months, study termination |
| Sendai | 6, 12, 14, and 17 months, study termination |
| Immunofluorescence Assay | |
| RCV/SDA | Study termination |
| Hemagglutination Inhibition | |
| H-1 | 6, 12, 14, and 17 months, study termination |
| KRV | 6, 12, 14, and 17 months, study termination |

## RESULTS

One rat had a positive titer to *M. arthritidis* at the end of the 2-year study. Further evaluation of the sample positive for *M. arthritidis* by immunoblot and Western blot procedures indicated that the positive titer may have been due to a cross reaction with antibodies of nonpathogenic *Mycoplasma* or other agents. There were no clinical findings or histopathologic changes of *M. arthritidis* infection in the rat with the positive titer. Accordingly, the *M. arthritidis*-positive titer was considered to be a false positive.

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