NTP REPORT ON THE

TOXICITY STUDIES OF ANTIMONY POTASSIUM TARTRATE IN F344/N RATS AND B6C3F$_1$ MICE

(DRINKING WATER AND INTRAPERITONEAL INJECTION STUDIES)

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

March 1992

NTP TOX 11

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U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
Public Health Service
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FOREWARD

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 the 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 toxicity study report were performed under the direction of the NIEHS and were conducted in compliance with NTP chemical health and safety requirements and must meet or exceed all applicable federal, state, and local health and safety regulations. Animal care and use were in accordance with the Public Health Service Policy on Humane Care and Use of Animals.

Anyone who is aware of related ongoing or published studies not mentioned in this report, or of any errors in this report, is encouraged to make this information known to the NTP. Comments and questions should be directed to Dr. J.R. Bucher, NIEHS, P.O. Box 12233, Research Triangle Park NC 27709 (919) 541-4532).

These studies are designed and conducted to characterize and evaluate the toxicologic potential of selected chemicals in laboratory animals (usually two species, rats and mice). Chemicals selected for NTP toxicology studies are chosen primarily on the bases of human exposure, level of production, and chemical structure. Selection per se is not an indicator of a chemical's toxic potential. Single copies of this Report are available without charge while supplies last from the NTP Public Information Office, NIEHS, P.O. Box 12233, Research Triangle Park, NC 27709 (919-541-3991).

TOXICITY STUDIES

OF

ANTIMONY POTASSIUM TARTRATE

(CAS NO.: 28300-74-5)

IN F344/N RATS AND B6C3F$_1$ MICE

(DRINKING WATER AND INTRAPERITONEAL INJECTION STUDIES)

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The NTP report on the toxicity studies of antimony potassium tartrate is based on the 14-day drinking water studies that began in December, 1986, and ended in January, 1987; the 16-day intraperitoneal injection studies that began in March, 1987, and ended in April, 1987; and the 13-week intraperitoneal injection studies that began in November, 1987, and ended in May, 1988.

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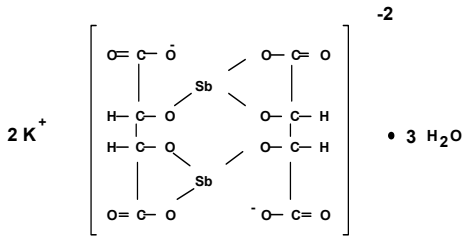
ANTIMONY POTASSIUM TARTRATE



CAS Number: 28300-74-5
Molecular formula: $C_8H_{10}K_2O_{15}Sb_2$
Molecular Weight: 667.8

Synonyms: APT; Tartar emetic; tartrated antimony; tartarized antimony; potassium antimonyltartrate; Bis [μ-[2,3-dihydroxybutanedioato(4-)-O1, O2:O3, O4]]-diantimonate dipotassium trihydrate (stereoisomer).

ABSTRACT

Antimony potassium tartrate (APT) is a complex salt that until recently was used worldwide as an anti-schistosomal drug. APT was efficacious in humans only if administered intravenously at a near-lethal total dose of 36 mg/kg. Because unconfirmed epidemiologic studies suggested a possible association between APT treatment and bladder cancer, prechronic toxicity studies were initiated with APT to select a route of administration and appropriate doses in the event chronic studies were needed. To determine the most appropriate route for longer-term studies, toxicity and concentrations of tissue antimony were compared in F344/N rats and $B6C3F_1$ mice that were administered APT in drinking water or by i.p. injection for 14 or 16 days. The animals were assigned to dose groups, 5/sex/species. Drinking water doses, estimated by water consumption, were 0, 16, 28, 59, 94, or 168 mg/kg in rats and 0, 59, 98, 174, 273, or 407 mg/kg in mice; i.p. doses were 0, 1.5, 3, 6, 11, or 22 mg/kg in rats and 0, 6, 13, 25, 50, or 100 mg/kg in mice.

APT was poorly absorbed and relatively nontoxic when given orally. There was no mortality or histopathological lesions in rats or mice receiving doses of APT as high as 168 or 273 mg/kg, respectively. One mouse in the highest dose group (407 mg/kg) died, and there were treatment-related lesions in the liver and forestomach of most mice in this dose group. In contrast, i.p. administration of the drug was much more toxic, resulting in the deaths of rats administered 22 mg/kg; kidney and liver lesions were found in these rats. In mice, i.p. administration of APT caused deaths and liver lesions at dose levels one-fourth of those that caused similar effects by oral administration. All male and female mice injected with 100 mg/kg APT died; half of the female mice given 50 mg/kg APT died; additional deaths occurred with doses as low as 6 mg/kg. Hepatocellular necrosis and inflammation of the liver capsule

were present in both sexes of mice in the 50 mg/kg dose groups. As a result of these findings, an i.p. dose regimen was selected for subsequent studies.

Groups of ten male and female F344/N rats and B6C3F$_1$ mice were given 0, 1.5, 3, 6, 12, or 24 mg/kg doses of APT 3 times per week for 13 weeks by i.p. injection. Rats were more sensitive than mice to the toxic effects of APT, exhibiting dose-related mortality and reduction in body weight. Four male rats in the 24 mg/kg dose died; body weights in both sexes of rats from this dose group and in male rats from the 12 mg/kg dose group were 10-20% below controls. No clinical signs of toxicity in the mice, nor gross or microscopic lesions, could be attributed to APT. Increased concentrations of antimony, considered to be dose-related, were detected in the blood, liver, kidney, spleen, and heart of rats, and in the liver and spleen of mice. In rats, hepatocellular degeneration and necrosis were associated with dose-related elevations in activities of the liver-specific serum enzymes, sorbitol dehydrogenase and alanine aminotransferase. By alternating the site of abdominal injection and the days of treatment, mesenteric inflammation at the site of administration was minimized in the rats and mice, indicating that the i.p. route would be suitable for chronic studies. Hepatotoxicity in rats occurred in dose groups where there was little evidence of renal toxicity and no cardiac toxicity; thus, serial measurement of liver-specific serum enzyme activities may be useful to monitor the presence and progression of hepatocellular degeneration in longer-term exposures.

PEER REVIEW PANEL

The members of the Peer Review Panel who evaluated the draft report on the toxicity studies on antimony potassium tartrate on November 20, 1990, are listed below. Panel members serve as independent scientists, not as representatives of any institution, company, or governmental agency. In this capacity, panel members act to determine if the design and conditions of the NTP studies were appropriate and to ensure that the toxicity studies report presents the experimental results and conclusions fully and clearly.

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SUMMARY OF PEER REVIEW COMMENTS ON THE TOXICOLOGY STUDIES OF ANTIMONY POTASSIUM TARTRATE

On November 20, 1990, the draft report on toxicity studies of antimony potassium tartrate received public review by the Technical Reports Review Subcommittee and associated Panel of Experts of the National Toxicology Program's Board of Scientific Counselors. The review meeting was held at the National Institute of Environmental Health Sciences in Research Triangle Park, North Carolina.

Dr. Michael Dieter, NIEHS, began the discussion by reviewing the design and results of the antimony potassium tartrate studies.

Dr. Klaassen, a principal reviewer, noted several suggested editorial changes. Dr. Carlson also noted editorial changes and asked if the program had considered performing genotoxicity assays in mammalian cells to extend the *Salmonella* studies currently reported. Dr. Zeiger responded that decisions on further genetic toxicity studies had not yet been made. Other panel members discussed further editorial matters; Dr. Scala then accepted the report on behalf of the panel.

I. INTRODUCTION

Antimony potassium tartrate (APT) is a complex salt that has been used as an intravenously administered anti-helminthic agent. APT has only a narrow margin of therapeutic safety, and cases of blindness, hydrocephalus, glomerulonephritis, myocardial lesions, and arrhythmia have been reported among the approximately 5 million people per year receiving this drug for the treatment of schistosomiasis (AMA, 1983).

APT acts to inhibit phosophofructokinase, blocking the obligate pathway for the anaerobic metabolism of glucose in *Schistosoma japonicum* (Bueding and Schiller, 1968; Saz and Bueding, 1966). A typical cumulative dose administered intravenously to humans over alternate days is 2500 mg (ca. 36 mg/kg in a 70 kg human), initiated at 40 mg per day and increased by 20 mg increments until 140 mg is reached (ca. 2 mg/kg), whereupon this dose is continued for the duration of the treatment regimen (AMA, 1983). Tolerance to the drug is variable in humans and is dependent on size, age, general health, severity of the parasitic infestation, etc. By comparison, rats and mice in the present studies were given a range of i.p. injections from 1.5 mg/kg up to 24 mg/kg 3 times per week for 13 weeks, which corresponds to a cumulative dose of approximately 250 mg in rats or 25 mg in mice (both ca. 1 g/kg) for the highest dose group.

The toxicity of APT, or tartar emetic, has been investigated since the early 1900's in rabbits (Meneghetti, 1941, and Lucia and Brown, 1941, both cited in Bradley and Fredrick, 1941; Oelkers 1947, Franz 1947, Pribyl, 1947, and Wieland, 1947, all cited in Fairhall and Hyslop, 1947); and in rats (Bradley and Fredrick, 1941; Van Esveld, 1947, cited in Fairhall and Hyslop, 1947). It was reported that doses in the lethal range caused toxicity to the formed elements in the blood, and to the heart, liver, kidney, and spleen of rabbits and rats. A review of the acute toxicity data showed that parenteral routes of administration (i.p., i.v., s.c.) resulted in toxic effects at doses about 1/10th those found toxic by oral administration (Venugopal and Luckey, 1978).

APT is slowly absorbed from the gastrointestinal tract, where it causes local irritation and sloughing of mucosal tissues, resulting in severe vomiting. Intravenously administered APT accumulates in erythrocytes, with lesser amounts distributed to other tissues, predominantly the liver, adrenals, spleen, and thyroid (Venugopal and Luckey, 1978). The metabolism of antimony was reviewed in the 1940's and again in the 1950's (Fairhall and Hyslop, 1947; Fairhall, 1957). Slow excretion, predominantly in the feces but also in the urine, followed ingestion of radiolabeled APT by rats (Weitz and Ober, 1965).

Epidemiologic studies suggested there was an association between lung tumors and antimony exposure in smelter workers (Davies, 1973). In rodent studies, antimony trioxide was reported to be carcinogenic when administered as particles by inhalation (Watt, 1983); similar findings were reported in a study of antimony trioxide or antimony ore concentrate (Groth *et al.*, 1986). There also was some concern about the potential carcinogenicity of APT, based on an unconfirmed association between drug treatment and bladder tumors in humans (El-Aser *et al.*, 1979; Kelada *et al.,* 1972). Three chronic carcinogenicity studies with APT were negative in rats and mice, but these studies were considered inadequate for assessment of carcinogenicity

because limitations in dose, route, sample number, and histopathology combined to reduce the power of the tests (Kanisawa and Schroeder, 1969; Schroeder *et al.*, 1968, 1970).

Chemicals containing antimony were negative for mutation induction in *Salmonella* assays, but were positive in mutagenicity tests with animal and human cells. The frequency of chromosomal aberrations (ABS) reportedly increased significantly in bone marrow cells of rats at 6, 24, and 48 hours after a single injection of 2.0-14.8 mg/kg antimony potassium tartrate (El Nahas *et al.*, 1982); treatment with these same doses once a day for 5 days (cells harvested 6 hours after final injection) also resulted in a statistically significant increase in ABS, although the response did not correlate with the dose. (The highest dose produced the smallest increase in ABS.) Results from similar experiments with the related chemical piperazine antimony tartrate (both single and multiple injections, 1.0-19.1 mg/kg) also demonstrated clastogenicity, but the response was less than that observed with antimony potassium tartrate and did not correlate with dose (El Nahas *et al.*, 1982). For the multiple injection protocol, the only significant increase in ABS occurred at the mid-dose. Antimony sodium tartrate, at a concentration of 2.3×10^{-9} M, produced a significant increase in chromosomal aberrations (ABS) in 48 hour cultures of human leukocytes (Paton and Allison, 1972).

Prechronic studies with APT were initiated in case additional epidemiological evidence became available to warrant further chronic studies. The drug was given either by i.p. injection or *ad libitum* in the drinking water to F344/N rats and $B6C3F_1$ mice for 14 or 16 days to compare toxicity and select the most appropriate route of administration for chronic studies. The data indicated that poor absorption would limit the usefulness of the oral route. Subsequent 13-week studies were conducted using i.p. injection 3 times per week on alternate sides of the abdomen, to minimize the potential for inflammation at the site of injection. The 13-week studies were performed to determine if this treatment regimen would be feasible in 2-year studies.

Wide ranges of APT doses were employed in the rodent studies because APT is administered at toxic doses for the treatment of parasitism in humans (AMA, 1983). Evaluation of the lower end of the toxicity curve would reveal the organs most sensitive to APT toxicity. In addition to examining animals for clinical signs and histopathological changes, measurements were taken of antimony accumulation in the blood, liver, kidney, spleen, and heart, of tissue-specific serum enzyme and isozyme, and of urinary enzyme responses.

II. MATERIALS AND METHODS

Procurement and Characterization of Antimony Potassium Tartrate

APT was purchased from Pfizer Chemical Div. (Hoffman Estates, IL). Analyses for identity and purity were performed by infrared spectroscopy and high performance liquid chromatography. The purity of the chemical was 99.4 - 100%; no breakdown occurred during storage.

For the 14-day studies, dose formulations of 0, 0.15, 0.30, 0.65, 1.25, and 2.5 mg/ml for rats, and 0, 0.30, 0.65, 1.25, 2.5, and 5.0 mg/ml for mice, were prepared in deionized water for *ad libitum* delivery in drinking water. Doses for the 16-day i.p. injection studies were prepared in physiological saline and were formulated at 0, 0.625, 1.25, 2.5, 5.0, and 10.0 mg/ml for both species. The dose solutions were stored in the dark at room temperature; analyses for APT concentrations, performed before and after administration to the animals, were found to be within 10 percent of their nominal concentrations.

For the 13-week i.p. injection studies, dose solutions were formulated in physiological saline at 0, 0.313, 0.625, 1.25, 2.5, and 5.0 mg/ml for mice, and at 0, 0.625, 1.25, 2.5, 5.0, and 10.0 mg/ml for rats. Doses were prepared every 2 weeks and were not used longer than 3 weeks. Dose mixture analyses performed before and after administration to the animals indicated APT concentrations within 10 percent of their nominal concentrations.

Animals

Six-week old male and female F344/N rats and $B6C3F_1$ mice were obtained from NTP animal breeding colonies from several suppliers (Simonsen Labs, Inc., Gilroy, CA; Taconic Farms, Inc., Germantown, NY; NCI-Frederick Cancer Research Facility, Frederick, MD) and held for 11-21 days before dose administration began. Animals were randomized to treatment groups; rats were housed 5 per cage, and mice were individually housed in polycarbonate, filter-top cages with hardwood bedding. They were given free access to Zeigler NIH-07 Open Formula diet (Zeigler Brothers, Inc., Gardners, PA). Animals were maintained in rooms on a 12-hour light/dark fluorescent light cycle, with a minimum of 10 room air changes per hour, at 21-24 C° in 35%-65% relative humidity.

14-Day Drinking Water Studies

Male and female F344/N rats and $B6C3F_1$ mice were given APT continuously in their drinking water for 14 days, followed by a 1-day observation period when animals were not dosed. Water consumption was measured at the end of each 7-day exposure period. The daily mg/kg dosages, calculated using the water consumption data and the body weight averages, were 0, 16, 28, 59, 94, or 168 mg/kg body weight in rats and 0, 59, 98, 174, 273, or 407 mg/kg body weight in mice. Groups of 5 animals per sex, species, and dose were evaluated histopathologically as outlined in Table 1. Antimony concentration was measured in the blood, kidney, heart, liver, and spleen of rats and mice (5 per group) at study termination, according to the methods described for the 13-week study.

16-Day Intraperitoneal Injection Studies

Male and female F344/N rats and B6C3F$_1$ mice were given 12 i.p. injections of APT over a 16-day period, administered on weekdays only. Dose volumes were based on each treatment group's most recent body weight, taken on day 1 and on day 7 of the studies. The dose volume given to control male and female animals equalled the greatest volume calculated for any of the treated groups of the respective sex. The daily mg/kg dosages, calculated using the body weight averages, were 0, 1.5, 3, 6, 11, or 22 mg/kg body weight in rats and 0, 6, 13, 25, 50, or 100 mg/kg body weight in mice. Groups of 5 animals per sex, species, and dose were evaluated histopathologically as outlined in Table 1. Antimony was measured in the blood, kidney, heart, liver, and spleen of rats and mice (5 per group) at study termination, according to the methods described for the 13-week study.

13-Week Intraperitoneal Injection Studies

The i.p. route of administration was selected for 13-week studies of APT because higher drug concentrations could be achieved in blood and tissues using lower doses, simulating intravenous drug treatment in humans. Dose selections were based on mortality, body weight differences, and histopathological responses in the 16-day studies, but the frequency of administration was changed. Three doses were given within each 1-week period for 13 weeks. Dose volumes were adjusted to body weight; injection volumes ranged from 0.3 ml at the study start to 0.9 ml at study termination.

Groups of 30 rats and mice per sex were given antimony potassium tartrate 3 times per week by intraperitoneal injection at doses of 0, 1.5, 3, 6, 12, or 24 mg/kg body weight; the control group was given the vehicle only (physiological saline). Groups of 10 animals/sex/species/dose were used for histopathologic evaluation, and groups of 20 animals were used in clinical pathology studies. After 45 and 90 days on test, 10 rats and mice/sex were chosen at random from the clinical pathology study groups for hematology, urinalysis, and serum and urinary enzyme analyses.

Clinical Examinations, Supplemental Studies, and Pathology

Details of clinical examinations and pathology procedures are outlined in Table 1. Animals surviving to the end of the studies were killed humanely by exposure to carbon dioxide. The liver, thymus, right kidney, right testis, spleen, heart, brain, and lungs were collected from the core study group and clinical pathology study group of rats and from the core study group of mice at study termination; weights were recorded to the nearest 1.0 mg.

Clinical pathology evaluations in the 13-week studies included urinalysis and hematology in rats and mice; serum clinical chemistry assays were performed in rats only. Urine samples were collected over a 16-hour period from animals maintained in metabolism cages, at least 48 hours prior to blood collection and termination for tissue collection. Blood for hematologic and serum enzyme analyses was obtained from the retroorbital sinus of rats and mice that had been anesthetized with a mixture of carbon dioxide and oxygen. Serum enzymes (rats only), urinary

enzymes, urinalysis, and complete hematology in rats and mice were measured at 45 or 47 days of treatment and at study termination.

Serum enzymes were measured on a Hitachi 704® chemistry analyzer (Hitachi Boehringer Mannheim Diagnostics, Inc., Indianapolis, IN) using the procedure by Gay *et al.* (1968) and Wacker *et al.* (1956) for lactate dehydrogenase; using a test kit from Sigma Chemical Co. (St. Louis, MO) with methods similar to those described by Asada and Galambos (1963) and Weismer *et al.* (1965) for sorbitol dehydrogenase; using the methods of Henry *et al.,* (1960), as described by Bergmeyer and Horder (1980), for alanine aminotransferase; using the methods of Hausamen *et al.* (1967) for alkaline phosphatase; and a method based upon modification of the Szasz method (1976) for creatine kinase. Isoenzymes for creatine kinase (HH, BB, HB creatine kinase isozymes) and lactate dehydrogenase (L_{1-5} lactate dehydrogenase isozymes) were quantitated after application to agarose gel and electrophoresis, using a Paragon® kit (Beckman Instruments, Inc., Brea, CA). Gel plates were scanned on a Cliniscan II® Densitometer (Helena Laboratories, Beaumont, TX). Total bilirubin was measured by the end point method based on the procedure by Wahlefeld *et al.* (1972).

Urinary enzymes were also measured on a Hitachi 704® chemistry analyzer using a method of Maruhn *et al.* (1976) for beta-glucuronidase. A method based on the work of Lockwood and Bosnan (1979) was used for N-acetyl glucosaminidase. An optimization of the Karmen method as described by Bergmeyer *et al.* (1978) was used for aspartate aminotransferase, and the method of Szasz *et al.* (1974) was used for gamma glutamyl transpeptidase. The assays of urinary aspartate aminotransferase, lactate dehydrogenase, and alkaline phosphatase were modified for urine by Chin and Kozbelt (1978). Urinary creatinine was assayed; urine volume, appearance, specific gravity, and microscopic examination also were evaluated as a measure of toxicity to the urinary tract in both species.

Hematology measurements included differential leukocyte, leukocyte, erythrocyte, reticulocyte, and platelet counts; hematocrit; methemoglobin measured by the method of Evelyn and Malloy as described by Henry (1974); hemoglobin concentration (HGB); mean corpuscular hemoglobin (MCH); mean corpuscular hemoglobin concentration (MCHC); and mean cell volume (MCV). Hematology data, except for reticulocyte and differential counts, were obtained by using an Ortho ELT-8 Hematology Analyzer (Ortho Diagnostic Systems, Raritan, NJ).

Analyses of blood and tissue antimony were measured by atomic absorption spectrophotometry of samples after acid digestion. Samples were presented by atomization in a matrix of 1% sulfuric acid:4-5% nitric acid. The analytical system was a Perkin-Elmer Zeeman/3030 Atomic Absorption Spectrophotometer, Perkin-Elmer HGA-600, and a Perkin-Elmer AS-60 with a pyrolytically coated graphite tube (Perkin-Elmer Co., Norwalk, CT). The parameters were set as a wavelength of 217.6 nm, a slit width of 0.7 nm, a sample volume of 10-30 µl, a matrix modifier/volume of Ni/5 µl, with a maximized furnace program based on sample volume. Recovery of antimony from representative tissue preparations was 87 ± 3%. The limits of detection for antimony were dependent on the type of sample, and in mice were 0.03 µg/g for liver and 0.70 µg/g for spleen; in rats the detection limits ranged from 0.10 to 0.40 µg/g for blood, liver, spleen, kidney, and heart.

A necropsy was performed on the core and the clinical pathology study groups of rats and on the core study group of mice. Organs and tissues were examined for gross lesions, and tissues were preserved in 10% neutral buffered formalin. Tissues routinely processed for preparation of histologic sections for microscopic examination are listed in Table 1.

Upon completion of the histologic evaluation by the laboratory pathologist, 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 sent to an independent pathology laboratory where quality assessment was performed, and the results were reviewed and evaluated by the NTP Pathology Working Group (PWG). Tissues evaluated by the PWG included the liver, kidney, and mesentery in rats and mesentery in mice. The final diagnoses represent a consensus of contractor pathologists and the PWG. Details of these review procedures have been described by Maronpot and Boorman (1982) and Boorman *et al.* (1985).

Virology Screen

Blood samples were collected and the sera analyzed for viral titers from 5 animals per sex and species at study start and termination in the 13-week studies. Data from 5 viral screens performed in rats and 12 viral screens performed in mice showed that there were no positive antibody titers (Boorman *et al.*, 1986; Rao *et al.,* 1989, 1989a).

Reproductive Toxicity

To screen for potential reproductive toxicity, sperm motility and morphology was evaluated at necropsy, and vaginal cytology was evaluated on the core study group of animals during the week just preceding necropsy, using procedures outlined by Morrissey *et al.* (1988). For the 12 days prior to sacrifice, females were subject to vaginal lavage with saline. The relative preponderance of leukocytes, nucleated epithelial cells, and large squamous epithelial cells were used to identify the stages of the estrual cycle.

Sperm motility was evaluated at necropsy as follows: The sperm that extruded from a small cut in the distal caudal epididymis were dispersed in solution, cover slipped, and counted. Two independent observers counted the number of moving and non-moving sperm in 5 fields of 30 sperm or less per field. After sperm-sampling for motility evaluation, the cauda was placed in phosphate buffered saline (PBS) and minced; the solution was mixed gently and heat-fixed at 65°C. Sperm density subsequently was determined using a hemocytometer.

To quantify spermatogenesis, the left testicular spermatid head count was determined by removing the tunica albuginea and homogenizing the testis in PBS containing 10% DMSO. Homogenization-resistant spermatid nuclei were enumerated using a hemocytometer.

Statistical Methods

The significance of differences between dosed and control groups was assessed using nonparametric multiple comparisons procedures designed to protect against false positive inferences. Either Dunn's test or Williams' modification of Shirley's multiple comparisons procedure was applied, based on the occurrence of a dose-related response in the data (Dunn, 1964; Shirley, 1977; Williams, 1986). If the P value from Jonckheere's test (Hollander and Wolfe, 1973) for a dose-related trend was greater than or equal 0.10, Dunn's test was used rather than Shirley's test. Tables for each individual parameter show the results of Shirley's or Dunn's test (reported at the 0.05 and 0.01 levels). The outlier test of Dixon and Massey (1951) was employed to detect extreme values.

Treatment effects for vaginal cytology were investigated by applying a multivariate analysis of variance (Morrison, 1976) to the transformed data to test for the simultaneous equality of measurements across dose levels.

Quality Assurance

The studies of antimony potassium tartrate were performed in compliance with FDA Good Laboratory Practices regulations (21 CFR 58). The Quality Assurance Unit of Battelle Columbus Laboratories performed audits and inspections of protocols, procedures, data, and reports throughout the conduct of the studies. The operations of the Quality Assurance Unit were monitored by the NTP.

Table 1 Experimental Design and Materials and Methods in the Intraperitoneal and Drinking Wa Studies of Antimony Potassium Tartrate

14-Day Drinking Water Studies	16-Day Intraperitoneal Studies	13-Week Intraperitoneal Studies
Dates of Studies, Dec. , 1986 - Jan. , 1987	March - April, 1987	Nov. , 1987 - May, 1988
Strain and Species F344/N rats; B6C3F$_1$ mice	F344/N rats; B6C3F$_1$ mice	F344/N rats; B6C3F$_1$ mice
Animal Source Simonsen Labs, Inc., Gilroy, CA	Simonsen Labs, Inc., Gilroy, CA	Mice--NCI-Frederick Cancer Research Facility, Frederick, MD Rats--Taconic Farms, Inc., Germantown, NY
Chemical Source Pfizer Chemical Division, Hoffman Estates, IL	Pfizer Chemical Division, Hoffman Estates, IL	Pfizer Chemical Division, Hoffman Estates, IL
Study Laboratory Battelle Memorial Institute, Columbus Division	Battelle Memorial Institute, Columbus Division	Battelle Memorial Institute, Columbus Division
Size of Study Groups Core study: 5/sex/group of each species. Special studies: 5/sex/group of each species. Rats were housed 5 per cage , and mice were individually caged.	Core study: 5/sex/group of each species. Special studies: 5/sex/group of each species. Rats were housed 5 per cage , and mice were individually caged.	30/sex/group of each species, (10 core study and 20 special study). Rats were housed 5 per cage and mice were individually caged.
Doses Mice--0, 0.3, 0.65, 1.25, 2.5, and 5.0 mg/ml; rats--0, 0.15, 0.3, 0.65, 1.25, and 2.5 mg/ml *ad libitum* in water.	Mice--0, 6, 13, 25, 50 and 100 mg APT/kg body weight; rats--0, 1.5, 3, 6, 11 and 22 mg APT/kg body weight in physiological saline.	Mice--0, 1.5, 3, 6, 12, and 24 mg APT/kg body weight; rats--0, 1.5, 3, 6, 12 and 24 mg APT/kg body weight administered 3 times per week in physiological saline.
Method of Animal Distribution Animals weighed and randomized (by partitioning algorithm) into groups by sex, assigned to cages, and cages assigned to dose groups.	Animals weighed and randomized (by partitioning algorithm) into groups by sex, assigned to cages, and cages assigned to dose groups.	Animals weight-randomized into groups by sex, assigned to cages, and cages assigned to dose groups.
Diet NIH 07; available *ad libitum*	NIH 07; available *ad libitum*	NIH 07; available *ad libitum*
Animal Room Environment Temp--69-75°F; relative humidity--35-65%; fluorescent light 12 h/d; 10 room air changes/h.	Temp--69-75°F; relative humidity--35-65%; fluorescent light 12 h/d; 10 room air changes/h.	Temp--68-76°F; relative humidity--23-87%; fluorescent light 12 h/d; 10 room air changes/h.
Time Held Before Study Rats--16-17 d, mice--20-21 d	Rats, mice--11-12 d	Rats--12-14 d, mice--13-15 d
Age When Placed on Study Rats--44-45 d; mice--48-49 d	40-41 d	6-7 wk
Age When Killed Rats--60-61 d; mice--64-65 d	56-57 d	19-20 wk
Type and Frequency of Observation Observed 2 x d; weighed at days 1 and 8 and at terminal sacrifice; water consumption measured at days 7 or 8 and on day 15.	Observed 2 x d; weighed at days 1 and 8 and 7 (base and special group mice) or 1 and 2 and 8 and 7 (base and special group rats) and at terminal sacrifice.	Observed 2 x d; weighed weekly and at terminal sacrifice.

Table 1 Experimental Design and Materials and Methods in the Intraperitoneal and Drinking Water Studies of Antimony Potassium Tartrate (continued)

Necropsy and Histologic Examinations (13-week intraperitoneal studies)

Necropsy performed; tissues were preserved in 10% neutral buffered formalin, embedded in paraffin, sectioned, and stained with H&E for microscopic examination. The following tissues were examined microscopically from all high dose and control animals and all early deaths in other groups: adrenal gland, bone and bone marrow, brain, cecum, clitoral/ preputial glands, colon, duodenum, mesenteric lymph node, epididymis, esophagus, gall bladder (mice), heart, ileum, jejunum, kidney, lung, liver, mammary gland, mandibular lymph node, nasal cavity, ovary, pancreas, pituitary gland, prostate, parathyroid gland, rectum, salivary gland, skin, spleen, stomach, seminal vesicle, testis, thyroid, thymus, trachea, urinary bladder, uterus, and all gross lesions. In addition to gross lesions, the following tissues were examined in all other dosed groups: mesentery, liver, kidney (rats and mice, i.p. studies); liver and forestomach (mice, drinking water studies); kidney (rats, drinking water studies). Hematologic and serum chemical analyses performed; residues of antimony were measured in the blood, kidney, heart, liver, and spleen of rats and mice at study termination; sperm morphology and vaginal cytology evaluated in 10 male and female rats and mice from the control, 3, 6, and 12 mg/kg dose groups (13-week studies. Blood smears were made for erythrocyte micronuclei determinations in mice (13-week studies).

III. RESULTS

14-Day Drinking Water Studies in F334/N Rats

All rats given APT in drinking water survived to the end of the study (Table 2). Water consumption was reduced by 20%-30% in male rats receiving 94 and 168 mg/kg APT, and proportional reductions in water consumption from 10%-40% were observed in female rats dosed at 28, 59, 94, or 168 mg/kg APT. No clinical signs were detected that were related to APT treatment. Increases in relative liver weight occurred in male and female rats that were given the highest dose of APT; in addition, there was an increase in relative kidney weight in females from this dose group. There were no histopathological changes attributed to APT treatment, except in dosed male rats, where protein droplets normally present in the cytoplasm of the renal tubular epithelium were stained somewhat more prominently than in the kidney of control rats. There was no clear dose-response relationship in antimony concentrations in the tissues of rats dosed with 16, 59, or 94 mg/kg APT; blood levels of antimony ranged between 15 and 20 μg/gm and were about 3 times as high as levels in the kidney, heart, spleen, and liver (Figure 1a).

Table 2 Survival, Weight Gain, and Compound Consumption of F344/N Rats in the 14-Day Drinking Water Studies of Antimony Potassium Tartrate [a]

Concentration (mg/ml) in water	Compound consumption [b]	Survival [c]	Mean body weights (grams) Day 1	Day 8	Day 16	Final Weight Relative to Controls (%) [d]	Average Water Consumption (ml)
MALE							
0.00	0	10/10	122	154	190		17.4
0.15	16	10/10	122	154	194	102	18.0
0.30	28	10/10	124	156	192	101	15.9
0.65	59	10/10	122	154	193	102	16.3
1.25	94	10/10	122	149	188	99	13.4
2.50	168	10/10	122	143	184	97	11.9
FEMALE							
0.00	0	10/10	103	120	140		16.0
0.15	16	10/10	104	121	139	99	15.6
0.30	28	10/10	103	119	138	99	13.9
0.65	59	10/10	103	118	131	94	12.6
1.25	94	10/10	102	118	140	100	11.0
2.50	168	10/10	104	115	137	98	9.5

a Includes animals from histopathology and clinical pathology study groups.
b Mg/kg/day, based on average water consumption and body weights of male and female rat groups combined.
c Number surviving after 14 days/number of animals in dose group.
d (Dosed group mean/control group mean) x 100.

16-Day Intraperitoneal Injection Studies in F344/N Rats

APT was much more toxic by the i.p. route, causing mortality and histopathological lesions at doses about 1 order of magnitude below those achieved by the drinking water route. In the i.p. studies, 1 male and 2 female rats in the 22 mg/kg (highest) dose group died on day 2 of treatment (Table 3). There were minor decreases in body weight (Table 3) but no changes in organ weights (not shown).

Table 3 Survival and Weight Gain of F344/N Rats in the 16-Day Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	Survival [b]	Mean Body Weight (grams) Day 1	Day 8	Day 16	Final Weight Relative to Controls (%) [c]
MALE					
0.0	10/10	102	143	192	
1.5	10/10	102	145	195	102
3.0	9/10	102	145	194	101
6.0	10/10	102	144	189	98
11.0	10/10	104	143	185	96
22.0	9/10	102	142	179	93
FEMALE					
0.0	10/10	84	112	135	
1.5	10/10	86	112	135	100
3.0	10/10	84	111	133	99
6.0	10/10	85	111	133	99
11.0	10/10	88	112	133	99
22.0	8/10	82	105	127	94

a Includes animals from histopathology and clinical pathology groups.
b Number surviving at 16 days/number of animals per dose group.
c (Dosed group mean/control group mean) x 100.

Liver and/or kidney lesions were present in 1 male and 2 female rats from the 22 mg/kg high-dose group that died on the second day of the study (Table 4). Liver evidenced a mild to marked hepatocellular necrosis in the periportal portion of the lobule. In the kidney, a diffuse vacuolar degeneration (moderate severity) of the cytoplasm in the tubular epithelium was present in the outer stripe of the outer medulla. Inflammation of the mesentery was present in both treated and vehicle control rats. This generally consisted of the focal accumulation (minimal to moderate) of a mixed, inflammatory cell infiltrate in the mesentery, adjacent to the pancreas, mesenteric lymph node, or intestine. In a few rats, a focal area of degeneration or

Table 4 Histopathologic Lesions in F344/N Rats in the 16-day Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	0.0	1.5	3.0	6.0	11.0	22.0
Site/Lesion						
MALE						
Liver, necrosis	0	0	0	0	0	1
Kidney, degeneration	0	0	0	0	0	1
Mesentery, inflammation	3	2	1	3	3	4
FEMALE						
Liver, necrosis	0	0	0	0	0	2
Kidney, degeneration	0	0	0	0	0	1
Mesentery, inflammation	1	1	1	3	5	3

a 5 animals were examined in each group.

hemorrhage associated with inflammation was present in the mesenteric fat. There was no clear, dose-related increase in incidence or severity of mesenteric inflammation.

There was a clear, dose-related increase in antimony concentration in the blood, liver, spleen, heart, and kidney of rats given APT by i.p. injection, in contrast to the lack of dose-response when the chemical was administered in the drinking water (Figs. 1a, 1b). However, generally equivalent maximal concentrations of antimony were attained in the tissues of the rats, when they were given the highest dose of APT by either route.

13-Week Intraperitoneal Injection Studies in F344/N Rats

In the rats given APT injections for 13 weeks, 4 animals from the 24 mg/kg (high-dose) group died during the course of treatment (Table 5). Body weights in the 24 mg/kg group of male and female rats were significantly depressed throughout the study, with final weights 11-18% below those of controls (Figure 2). Male rats dosed with 12 mg/kg APT exhibited final body weights 8% below controls.

Table 5 Survival and Weight Gain of F344/N Rats in the 13-Week Intraperitoneal Injection Studies of Antimony Potassium Tartrate

Dose Concentration(mg/kg)	Survival [a]	Mean Body Weight (grams)			Final Weight Relative to Controls (%) [c]
		Initial	Final	Change [b]	
MALE					
0.0	10/10	158	376	218	
1.5	10/10	167	384	217	102.1
3.0	10/10	169	375	206	99.7
6.0	10/10	167	380	214	101.1
12.0	10/10	158	346	188	92.0
24.0	6/10	151	309	161	82.2
FEMALE					
0.0	10/10	131	228	96	
1.5	10/10	132	219	87	96.0
3.0	10/10	126	220	94	96.5
6.0	10/10	121	213	92	93.4
12.0	10/10	132	222	89	97.4
24.0	10/10	126	203	77	89.0

a Number surviving at 13 weeks/number of animals per dose group, includes only core-study rats.
b Mean weight change of the animals in each dose group.
c (Dosed group mean/Control group mean) x 100.

Liver weights (absolute and relative) were increased in both sexes of dosed rats; a slight increase in kidney weight was present in dosed female rats (See Appendix A, Table A1). Variable increases and decreases in weights of spleen, thymus, testis, and heart were seen in both sexes of dosed rats. These changes were most often a relative increase in organ weight, attributable to decreased body weight gain in dosed rats.

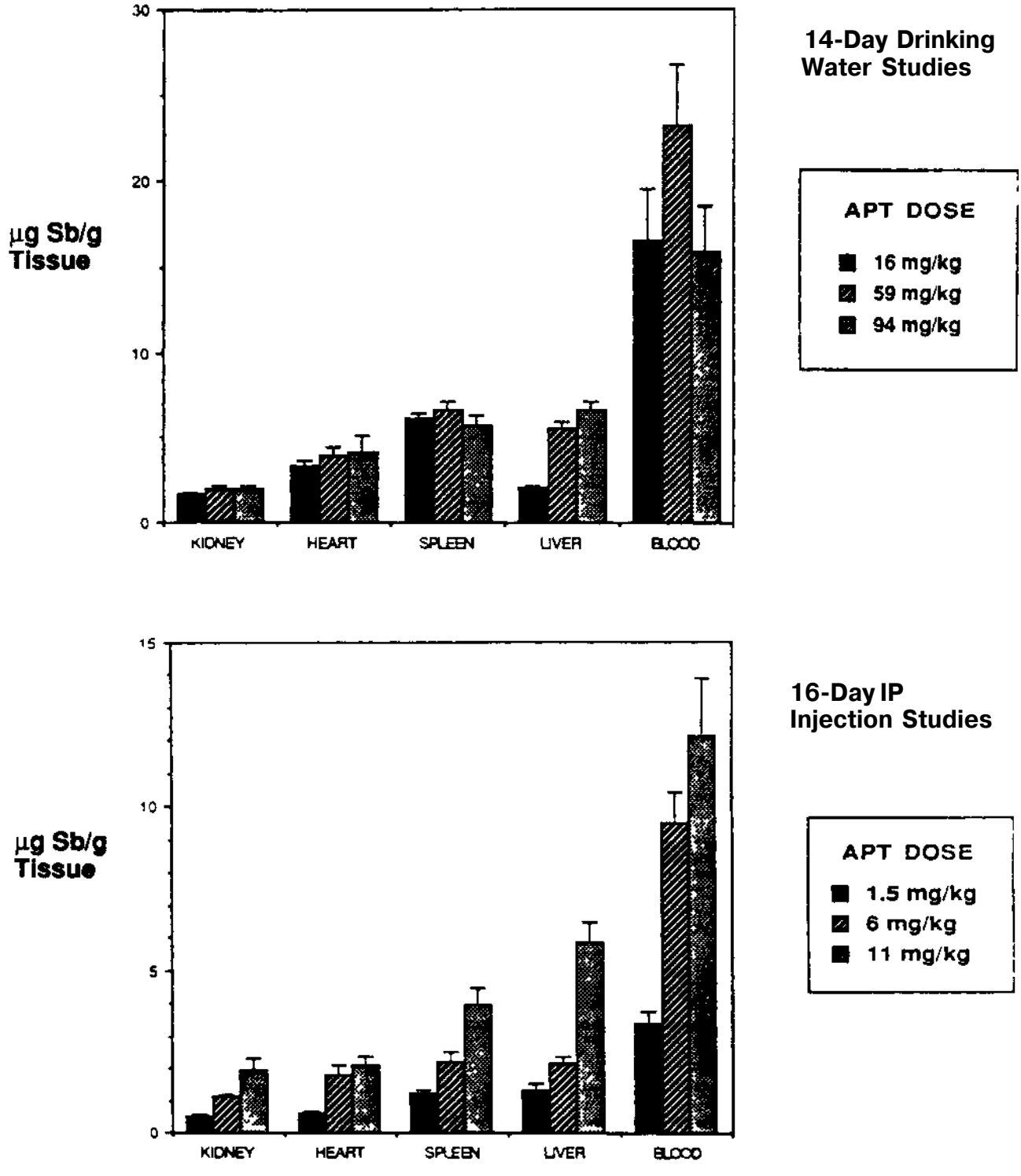

Figure 1 Concentrations of Antimony in F344/N Rat Tissues

Figures 1a and 1b. Concentrations of antimony in rat tissues after 14 days administration of APT in the drinking water or by intraperitoneal injection. There was no difference between sexes, and values were combined. Means ± SEM, N=10.

At study termination, antimony in the blood of male and female rats exhibited concentrations that were proportional to dose with 40, 80, and 100 µg/g levels in the 6, 12, and 24 mg/kg dose groups (Figure 3). The highest tissue levels of antimony were measured in the spleen (50 µg/g). Maximal antimony concentrations attained in the liver were about 30 µg/g tissue, with lower levels in the kidney (15 µg/g) and heart (10 µg/g).

There were no changes in hematological parameters in the treated rats, except for a dose-related decrease in the percentage and number of lymphocytes in male rats at study termination (Appendix B, Tables B1, B2).

The liver was the most sensitive of the tissues to antimony toxicity, judging from the responses in clinical and anatomic pathology. Among a panel of enzymes and isoenzymes in the serum and urine that are considered to be biological markers for liver, kidney, and heart toxicity, only those specific for liver were affected. The activities of serum alanine aminotransferase and sorbitol dehydrogenase in male and female rats increased significantly in proportion to dose and time after 45-47 days, or 94 days, of APT treatment (Figs. 4, 5). Total bilirubin concentrations also were elevated significantly in both sexes in the 24 mg/kg high-dose group. In contrast, there was no effect of APT treatment on lactate dehydrogenase, or on creatine kinase enzyme or isoenzyme activities that reflect cardiac toxicity; also, there was no change in urinary creatinine, or in the activities of a panel of 6 urinary enzymes that reflect tubular cell necrosis in the kidney (Appendix B, Tables B1, B2).

Table 6 Histopathologic Lesions in F344/N Rats in the 13-Week Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	0.0	1.5	3.0	6.0	12.0	24.0
Site/Lesion						
MALE						
Liver, degeneration	0	0	0	0	0	5
Liver, necrosis	0	0	0	2	8	6
Liver, bile duct hyperplasia	0	0	0	0	0	5
Liver Capsule, inflammation/fibrosis	0	0	3	10	9	8
Kidney, degeneration	0	0	0	0	0	3
Mesentery, inflammation	7	10	9	10	10	8
FEMALE						
Liver, degeneration	0	0	0	0	0	1
Liver, necrosis	0	0	0	0	1	10
Liver, bile duct hyperplasia	0	0	0	0	0	2
Liver Capsule, inflammation/fibrosis	0	0	0	7	10	10
Mesentery, inflammation	7	6	9	9	10	10

a Ten animals were examined in each group; the liver and kidney of the 2 high-dose male rats were not evaluated due to autolysis.

Histopathological changes in the liver were consistent with the clinical pathology results (Table 6). Treatment-related lesions in the liver were similar, morphologically, in both sexes, but slightly more severe in males. These lesions consisted of inflammation of the capsule and multiple foci of hepatocellular degeneration, or necrosis. Inflammation of the capsule (serosal surface) of the liver consisted of fibrosis, accumulation of pigment (hemosiderin) in macrophages, necrosis of hepatocytes adjacent to the capsule, and a minimal inflammatory cell infiltrate. This capsular lesion was dose-related in incidence and severity and did not occur in controls or in the lower dose groups. Among the higher dose groups, there was a prominent layer (1-5 cells in thickness) of hepatocellular necrosis adjacent to the areas of capsular fibrosis and inflammation (Plate 1, Figures 1 and 2). This lesion sometimes affected most of the capsular surface of the liver lobe. In some rats, particularly those in the highest dose groups, the capsular lesion was more extensive on the left lateral lobe than on the median lobe; at the lower doses (3.0 and 6.0 mg/kg) the capsular inflammation was sometimes present only at the tip of the liver lobe (Plate 1; Figure 5). Foci of degeneration and necrosis were sometimes periportal but usually distributed randomly throughout the hepatic lobules (Plate 1, Figures 3 and 4). Degeneration consisted of foci of swollen hepatocytes with pale, vacuolated cytoplasm. Inflammatory cell infiltrates were associated with foci of hepatocellular necrosis. At the highest dose (24 mg/kg), hyperplasia of bile ducts, periportal fibrosis, and accumulation of pigment (hemosiderin) within the cytoplasm of macrophages in portal areas were present. Fibrosis was more prominent in portal areas near the capsular surface.

Treatment-related degeneration of the kidney was present in 3 high-dose male rats. Tubular epithelial cells of the outer stripe of the outer medulla were swollen; the cytoplasm was vacuolated and there was necrosis of some cells. In 2 of the rats that survived until the scheduled sacrifice, there was also tubular cell regeneration in the outer stripe of the outer medulla. These findings were not reflected in the urinalyses or in increases in urinary enzyme markers for tubular cell injury.

Inflammation (minimal-to-mild) in the mesentery was present in all dose groups and vehicle control groups of male and female rats. The incidence and severity of inflammation were not dose-related. This lesion was distinctly different from the capsular inflammation in the liver. Mesenteric inflammation consisted of focal infiltrates of mononuclear and polymorphonuclear inflammatory cells in the mesentery/peritoneum attached to the pancreas, intestine, or mesenteric lymph node. This appeared to be a result of the intraperitoneal injections (Plate 1, Figure 6).

APT did not affect sperm morphology or the length of the estrous cycle in the reproductive toxicity screen of male and female rats (Appendix C, Tables C1, C2).

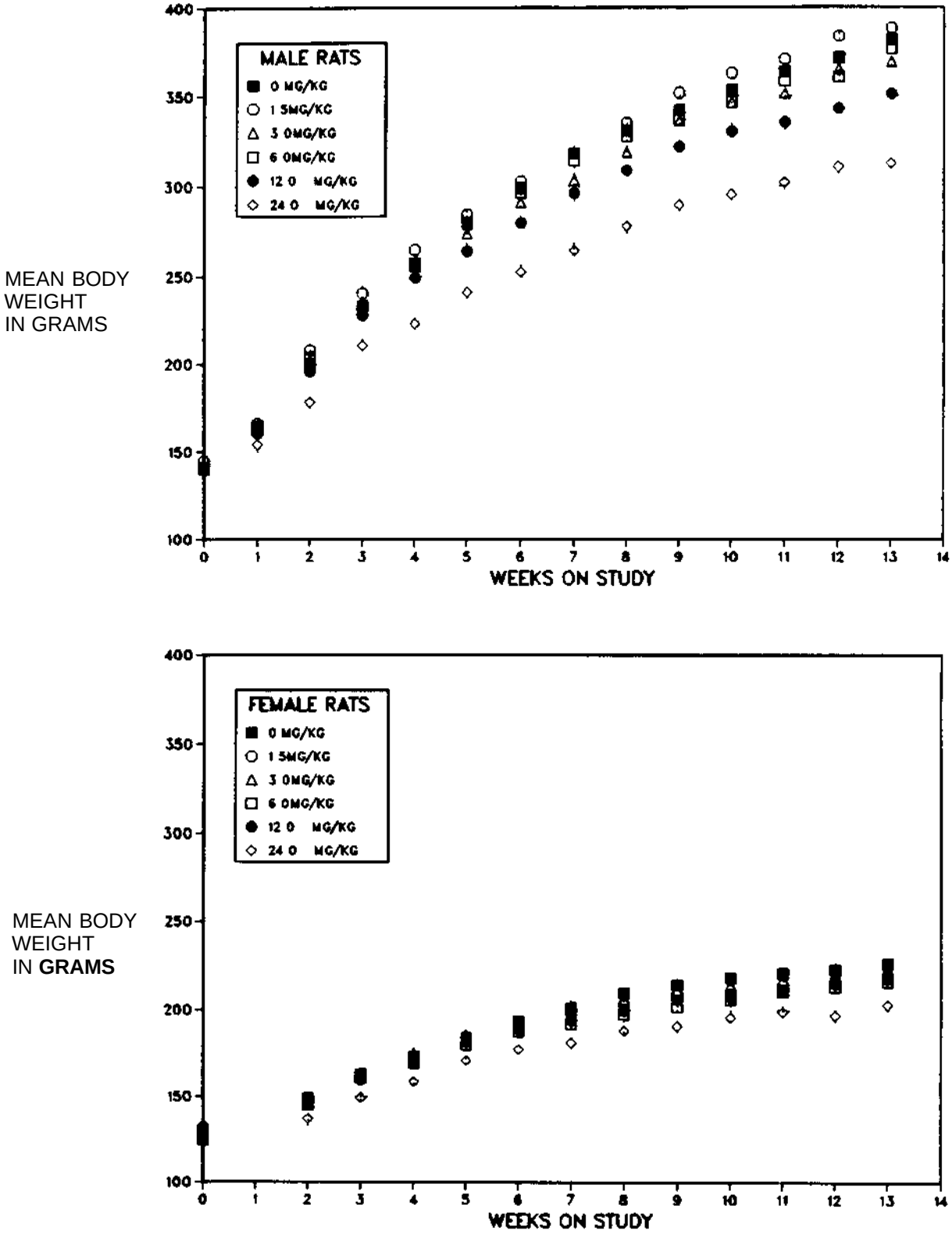

Figure 2 **Body Weights of F344/N Rats Exposed to Antimony Potassium Tartrate by Intraperitoneal Injection for 13 Weeks**

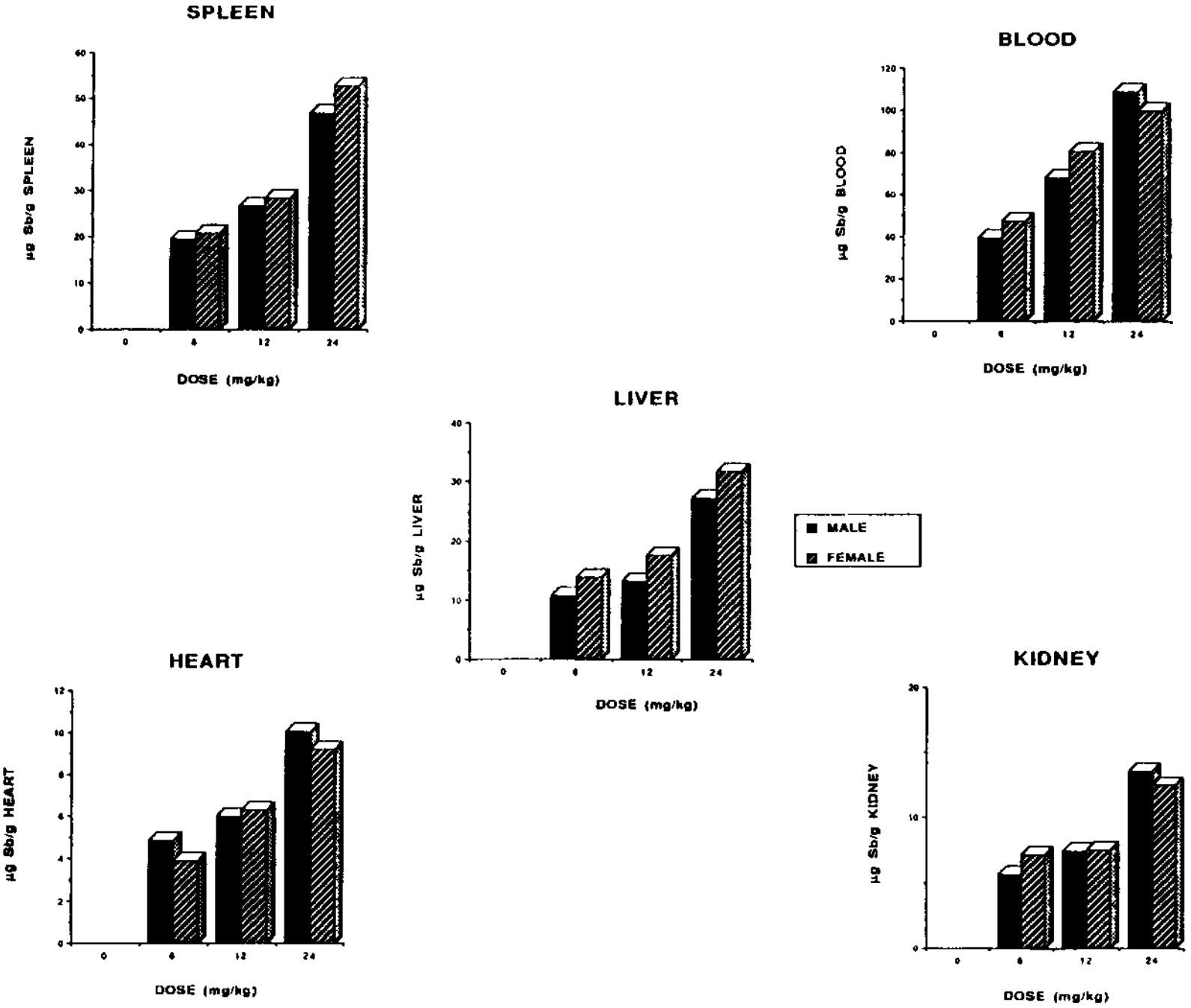

Figure 3 Concentrations of Antimony Potassium Tartrate in Tissues of F344/N Rats in the 13-Week Intraperitoneal Injection Studies

APT administered by intraperitoneal injection 3 days per week on alternate weekdays for 13 weeks Mean values, SEM≤10% of mean N=20 Significant increases in mid and high-dose above low-dose groups significant dose response trends

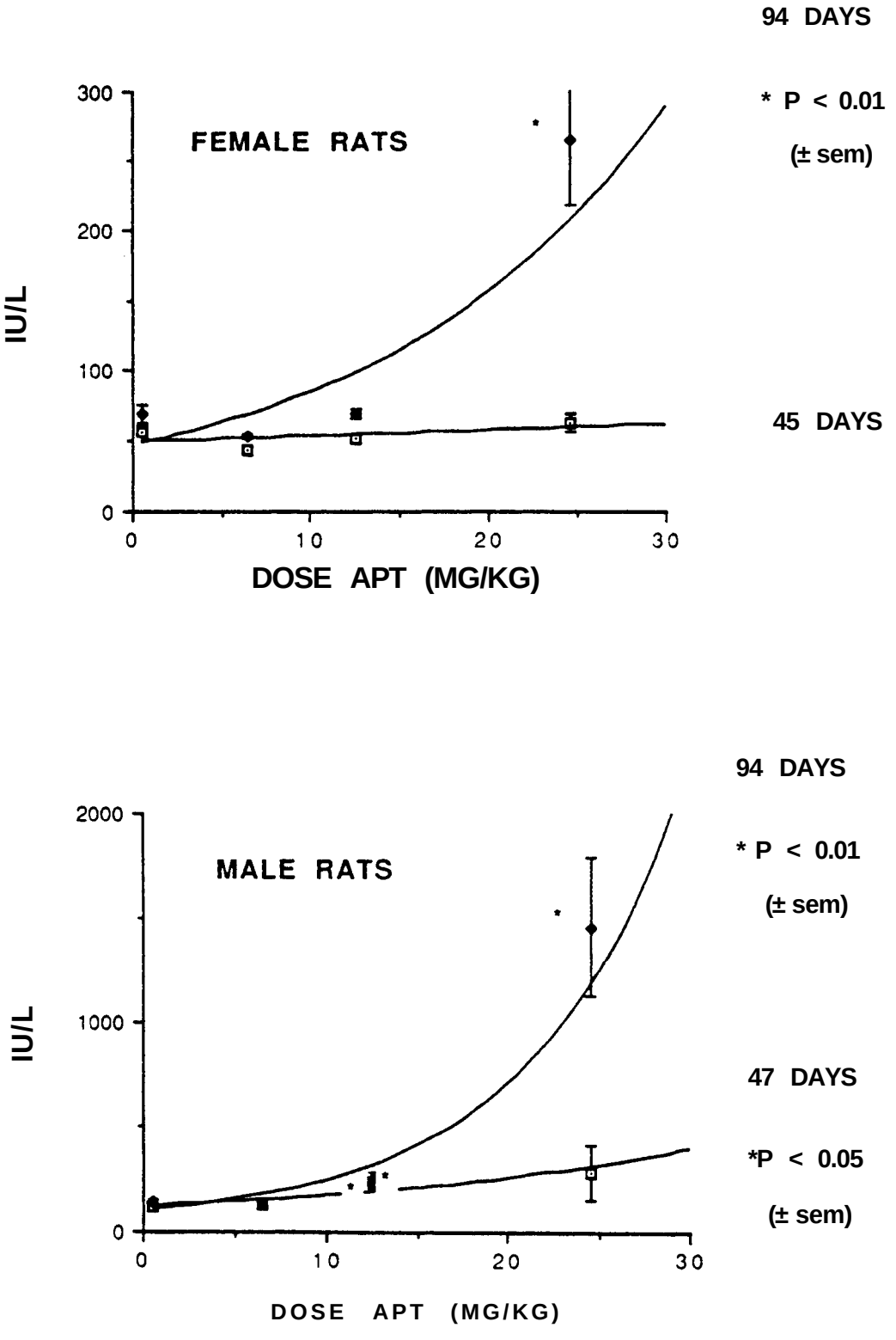

Figure 4 Serum Alanlne Amlnotransferase Activity in F344/N Rats Administered Antimony Potassium Tartrate by Intraperltoneal Injection for 13 Weeks

Serum alanine aminotransferase activity after administration of APT for 45 to 47 or 94 days. Injections administered 3 days per week on alternate weekdays. Means; N=10; best line fit for data calculated by regression.

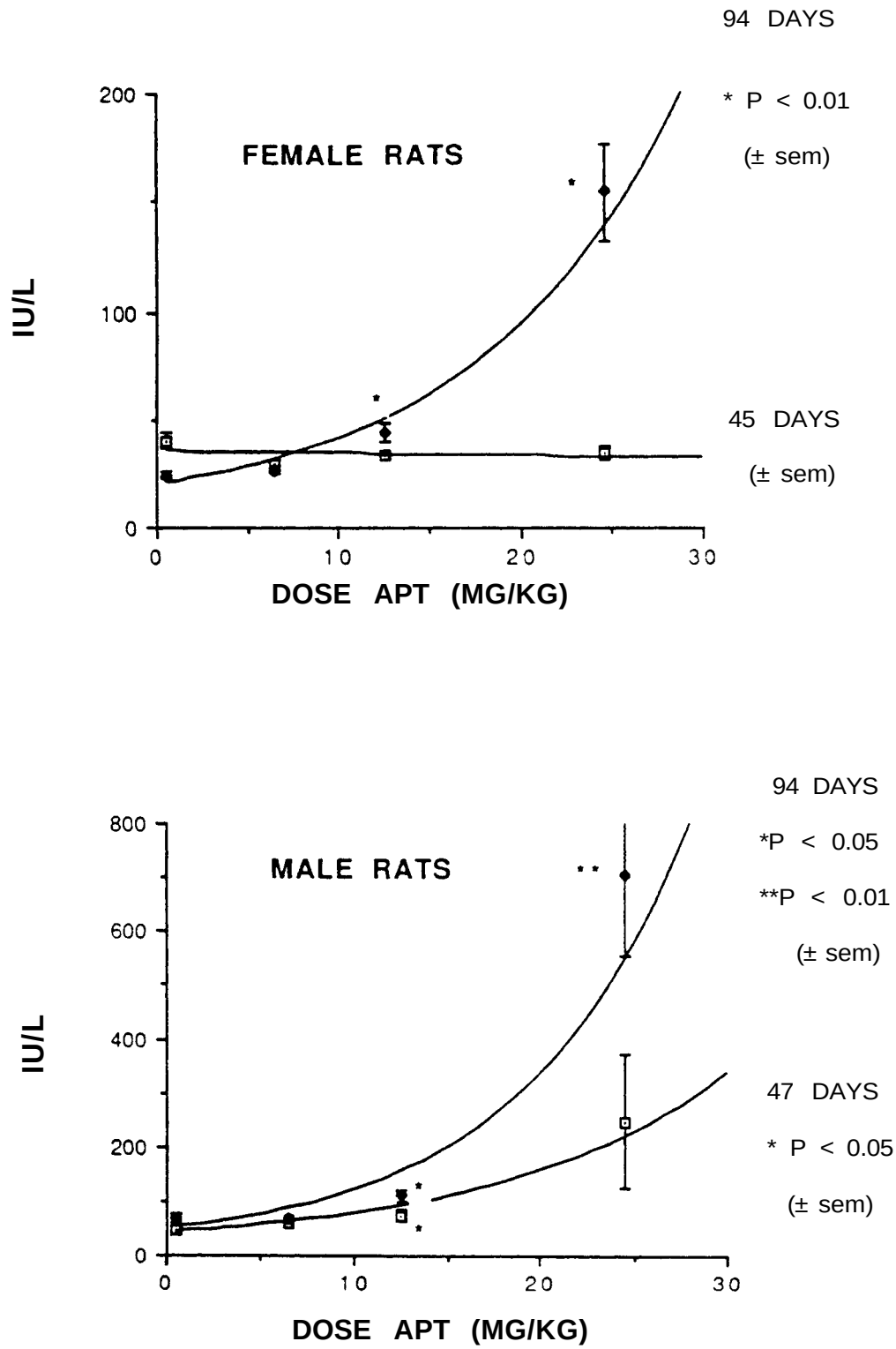

Figure 5 Sorbitol Dehydrogenase Activity in F344/N Rats Administered Antimony Potassium Tartrate by intraperitoneal Injection for 13 Weeks

Serum sorbitol dehydrogenase activity after administration of APT for 45 to 47 or 94 days. Injections administered 3 days per week on alternate weekdays. Means; N-10; best line fit for data calculated by regression.

PLATE 1

Histopathological Responses in F344/N Rats Administered Antimony Potassium Tartrate by Intraperitoneal Injection for 13 Weeks

Figure 1. Liver from male rat administered 24 mg/kg antimony potassium tartrate by the intraperitoneal route. Zone of hepatocellular necrosis and inflammation (arrows) along capsular surface.

Figure 2. Higher magnification of capsular lesion from Figure 1 shows zone of pale staining necrotic hepatocytes (N) and a minimal inflammatory cell infiltrate.

Figure 3. Liver from male rat administered 24 mg/kg antimony potassium tartrate by the intraperitoneal route shows multiple foci (arrows) consisting of hepatocellular necrosis with inflammation and congestion.

Figure 4. Higher magnification of a typical liver lesion shows focal hepatocellular necrosis and degeneration (pale staining cells) with inflammation and congestion of hepatic sinusoids.

Figure 5. Liver from female rat administered 6 mg/kg antimony potassium tartrate by the intraperitoneal route. Capsular lesion is limited to tip of lobe and consists of focal area of hepatocellular necrosis (N) and a mild inflammatory cell infiltrate (arrows).

Figure 6. Mesenteric attachment to small intestine (I) of rat administered 12.0 mg/kg antimony potassium tartrate by the intraperitoneal route. Minimal focal inflammatory cell infiltrate (arrows) present between fat cells of the mesentery.

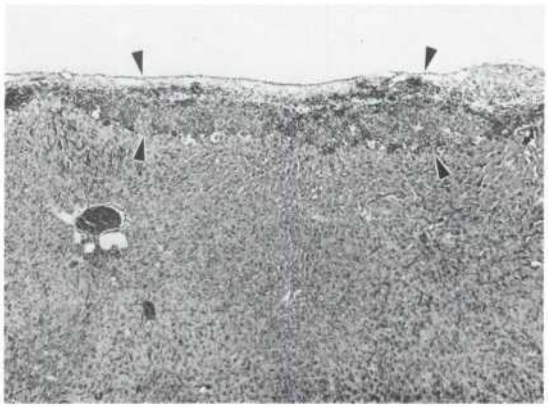

Figure 1

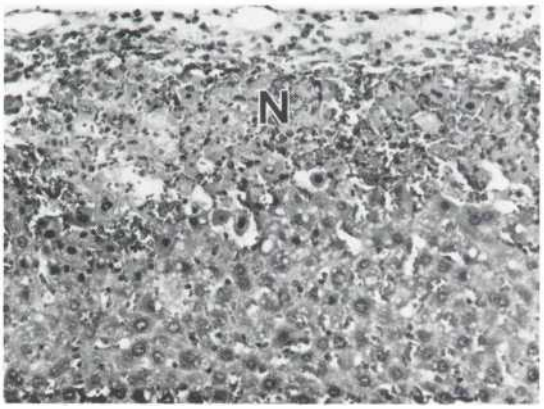

Figure 2

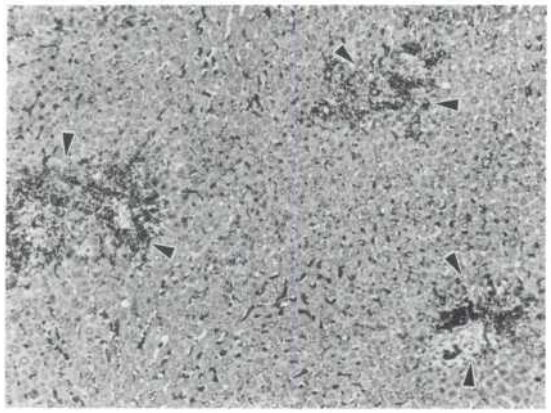

Figure 3

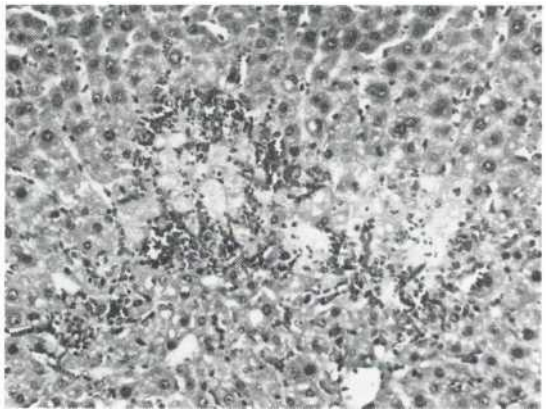

Figure 4

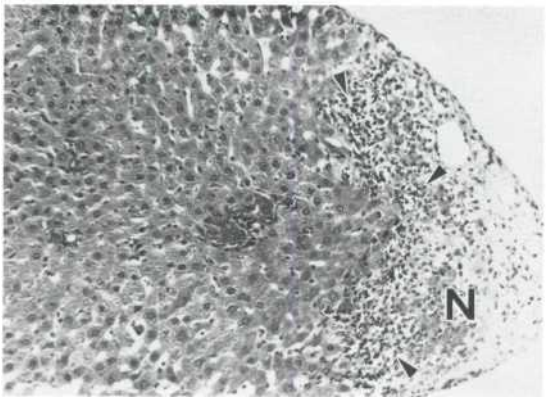

Figure 5

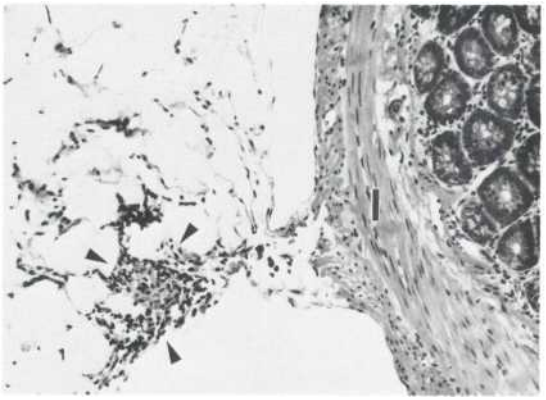

Figure 6

14-Day Drinking Water Studies in B6C3F$_1$ Mice

Among mice given APT in the drinking water, 1 female mouse in the 407 mg/kg (high-dose) group died (Table 7). Body weight gain was significantly reduced in males dosed with 273 or 407 mg/kg and in females dosed with 407 mg/kg APT for 8 days; however, recovery had occurred by day 16, except in the high-dose males (Table 7). Water consumption was apparently reduced in all treatment groups in proportion to dosage, but extreme variability in measurements limited the usefulness of the data. Clinical signs of toxicity included rough haircoat, emaciation, abnormal posture, hypoactivity, and decreased fecal material, consistent with avoidance of the APT-dosed water. Except for some dose-related increases in relative liver weight, no other biologically significant changes occurred in organ weights (not shown).

Table 7 Survival, Weight Gain, and Water Consumption of B6C3F$_1$ Mice in the 14-Day Drinking Water Studies of Antimony Potassium Tartrate [a]

Concentration (mg/ml) in water	Compound consumption [b]	Survival [c]	Mean Body Weight (grams) Day 1	Day 8	Day 16	Final Weight Relative to Controls (%) [d]	Average Water Consumption (ml/d)
MALE							
0	0	10/10	27	28	30		7.1
0.3	59	10/10	26	28	30	100	5.9
0.65	98	10/10	27	28	30	100	4.5
1.25	174	10/10	27	27	30	100	3.7
2.5	273	10/10	27	26	29	97	2.7
5.0	407	10/10	26	21	28	93	2.1
FEMALE							
0	0	10/10	20	22	24		14.1
0.3	59	10/10	21	22	24	100	7.6
0.65	98	10/10	21	22	24	100	4.9
1.25	174	10/10	21	22	24	100	3.7
2.5	273	10/10	21	21	24	100	3.0
5.0	407	9/10	20	18	24	100	2.1

a Includes animals from core and special study groups.
b Mg/kg/day, based on average water consumption and body weights of male and female mouse groups combined.
c Number of animals surviving at 14 days/number per dose group.
d (Dosed group mean/control group mean) x 100.

Treatment-related lesions were limited to the liver and forestomach in male and female mice in the highest dosage group (407 mg/kg). Gross lesions in the forestomach mucosa in 3 females and 1 male were composed of round, white nodules from 1 to 5 mm at their greatest dimension; 2 such nodules were present in the forestomach of 1 female. Microscopically, the nodules consisted of focal areas of ulceration with necrosis and inflammation of the squamous mucosa which extended into the underlying muscularis of the forestomach. Focal hyperplasia of the forestomach squamous epithelium was also associated with the areas of inflammation. There were no other microscopic lesions present in other portions of forestomach or glandular stomach. A minimal-to-moderate cytoplasmic vacuolization of hepatocytes was present in all male and female mice in the highest-dosed group (407 mg/kg); this lesion was not seen in lower dose groups or in controls. The cytoplasmic vacuolization was centrilobular in distribution but extended to the portal areas in some male mice, which generally showed more severe changes than females. Cells in the centrilobular region were slightly enlarged and

showed loss of cytoplasmic staining; nuclei were not displaced from their usual central location within hepatocytes.

16-Day Intraperitoneal Injection Studies in B6C3F$_1$ Mice

Administration of APT by the i.p. route resulted in deaths of all male and female mice at the highest dose group (100 mg/kg) during the first 2 days of treatment; 5/10 female mice in the 50 mg/kg dose group died by day 4. Thereafter, 1 female in the 6 mg/kg group and 3 in the 13 mg/kg also died, as did 1 male in the 13 mg/kg group and 1 male in the 25 mg/kg group (Table 8). There was no change in body weights (Table 8); the only change in organ weights was a decrease in lung weights in males and an increase in spleen weights in females in the 50 mg/kg dose groups (data not shown).

Table 8 Survival and Weight Gain of B6C3F$_1$ Mice in the 16-Day Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	Survival [b]	Mean Body Weight (grams) Day 1	Day 8	Day 16	Final Weight Relative to Controls (%) [c]
MALE					
0	10/10	23	24	25	
6	10/10	23	24	25	100
13	9/10	23	24	25	100
25	9/10	23	24	25	100
50	10/10	23	23	24	96
100	0/10	23	ND	ND	ND
FEMALE					
0	10/10	20	20	21	
6	9/10	20	20	23	110
13	7/10	20	20	21	100
25	10/10	19	20	22	105
50	5/10	19	19	22	105
100	0/10	20	ND	ND	ND

a Includes animals from core and special study groups.
b Number of animals surviving at 16 days/number per dose group.
c (Dosed group mean/Control group mean) x 100.
ND No data; all animals in group died before scheduled termination.

Microscopic lesions, attributed to the toxicity of APT administered by i.p. injection, were present in the liver of male and female mice from the 50 mg/kg dose group; there were no treatment-related lesions in the mice given 100 mg/kg which died within the first 2 days of the study (Table 9). Hepatocellular necrosis (minimal to mild) and inflammation (minimal) of the liver capsule was present in 4 male mice from the 50 mg/kg dose group. These lesions were characterized by individual necrosis of hepatocytes in the periportal areas; there were focal areas of fibrosis, inflammation, and mineralization in the liver capsule. Similar lesions were present in the liver of 2 female mice which died by the fourth day of the study and in a female which survived until the scheduled termination for the study. Inflammation in the mesentery was present in both treated and vehicle control mice. This consisted of the focal accumulation (minimal to moderate) of a mixed inflammatory cell infiltrate in the mesentery attached to the pancreas, mesenteric lymph node, or intestine. In several mice, a focal area of

fat necrosis was present with this inflammation. There was no clear dose-related effect with respect to incidence or severity of the inflammatory lesions in treated and vehicle control mice.

Table 9 Histopathologic Lesions in B6C3F$_1$ Mice in the 16-day Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	0	6	13	25	50	100 [b]
Site/Lesion						
MALE						
Liver, necrosis	0	0	0	0	4	0
Liver capsule, inflammation/fibrosis	0	0	0	0	4	0
Mesentery, inflammation	1	2	2	3	4	0
FEMALE						
Liver, necrosis	0	0	0	0	3	0
Liver capsule, Inflammation/fibrosis	0	0	0	0	1	0
Mesentery inflammation	1	3	1	3	1	0

a 5 animals were examined in each group unless otherwise specified.
b No animals in this group survived to the end of the study. None of the indicated lesions were found in the early death mice.

In mice, no antimony residues were detected in the blood, heart, or kidney; concentrations of approximately 24 µg/gm were detected in liver after 273 mg/kg APT was given in the drinking water or 50 mg/kg was given by i.p. injection; approximately 5 µg/gm was detected in spleens of mice given 50 mg/kg by i.p. injection.

13-Week Intraperitoneal Injection Studies in B6C3F$_1$ Mice

There was no mortality in female mice; the cause of the single deaths in the male mice control group and mid-dose groups could not be determined (Table 10).

There were no significant changes in body weight (Figure 6) and no clinical signs, nor were gross or microscopic lesions found that could be attributed to chemical treatment. There was minimal-to-mild inflammation of the mesentery in both the control and dosed groups, which was attributed to the route of administration (Table 11). Inflammation consisted of a focal infiltration of mononuclear and polymorphonuclear inflammatory cells in the mesentery/peritoneum of the pancreas, intestine, or mesenteric lymph node. There was no difference in incidence or severity which could be attributed to APT at doses up to 24 mg/kg administered 3 times per week by the intraperitoneal route.

There were reductions in red blood cell counts and hemoglobin in the both sexes of mice in the high dose groups at week 7 and again at week 13 in red cell counts. Relative spleen weights were increased in male mice at week 13 (Appendix A, Table A2).

The analyses of antimony concentrations in mice were restricted to the liver and spleen, since there were no detectable residues in the blood, kidney, and heart in the 14-day studies.

Antimony concentrations increased in a dose-related fashion in the liver of both sexes, with average levels of 7, 14, and 22 mg/g in the 6, 12, and 24 mg/kg dose groups. At each respec-tive dose, the spleens of females accumulated about half as much antimony as did the livers, while in male mice only spleens from the high-dose group exhibited measurable concentrations (3 µg/g) of antimony.

Table 10 Survival and Weight Gain of B6C3F$_1$ Mice in the 13-Week Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	Survival [a]	Mean Body Weight (grams)			Final Weight Relative to Controls (%)
		Initial	Final	Change[b]	
MALE					
0.0	9/10	22.2	32.3	9.9	
1.5	10/10	22.4	31.4	9.0	97.2
3.0	9/10	22.6	31.3	8.8	96.9
6.0	10/10	22.4	31.9	9.5	98.8
12.0	10/10	22.5	31.7	9.3	98.1
24.0	10/10	21.5	30.3	8.8	93.8
FEMALE					
0.0	10/10	18.7	26.8	8.0	
1.5	10/10	18.6	27.0	8.4	100.7
3.0	10/10	19.2	27.7	8.5	103.4
6.0	10/10	18.8	27.9	9.1	104.1
12.0	10/10	18.4	26.4	8.0	98.5
24.0	10/10	18.1	25.8	7.7	96.3

a Number of animals surviving at 13 weeks/number per dose group.
b Mean weight change of the animals in each dose group.
c (Dosed group mean/control group mean) x 100.

Table 11 Histopathologic Lesions in B6C3F$_1$ Mice in the 13-Week Intraperitoneal Injection Studies of Antimony Potassium Tartrate [a]

Dose (mg/kg)	0	1.5	3.0	6.0	12.0	24.0
MALE						
Mesentery, inflammation	8	8	9	7	4	8
FEMALE						
Mesentery, inflammation	4	6	10	6	10	10

a Ten animals were examined in each group.

In reproductive tissue evaluations (Appendix C, Tables C3, C4), there were decreases in testicular weights in mice in the 12 and 24 mg/kg dose groups, but these changes were not dose-related and constituted the only response in males. APT treatment did not affect sperm count or motility, and there were no effects on the average estrous cycle length in females.

Genetic Toxicology

Antimony potassium tartrate (10-10000 mg/plate) was not mutagenic in *Salmonella typhimurium* strains TA100, TA1535, TA97, or TA98 when tested with a preincubation protocol in the presence and the absence of exogenous metabolic activation (induced rodent liver S9) (see Appendix D).

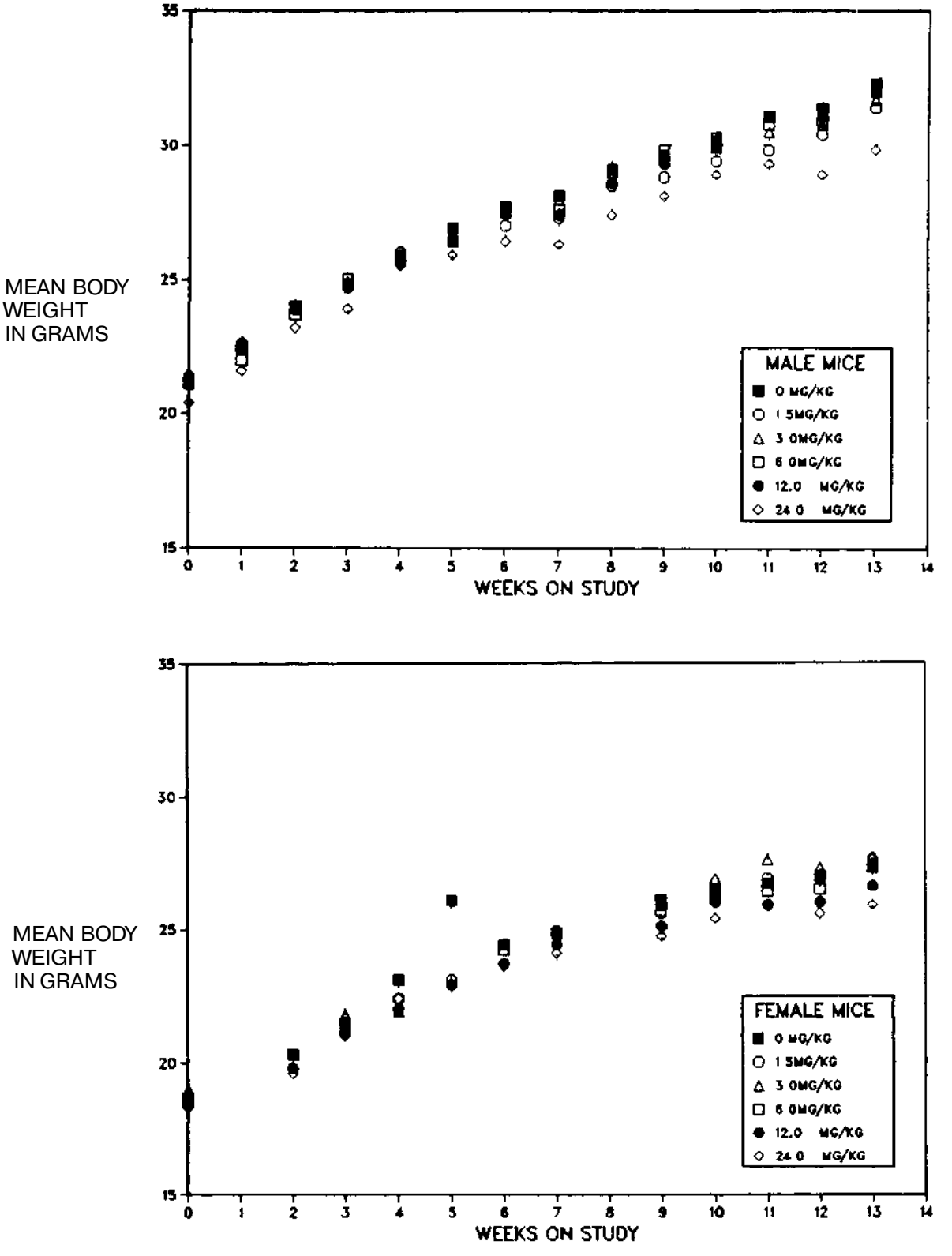

Figure 6 **Body Weights of B6C3F1 Mice Exposed to Antimony Potassium Tartrate by intraperitoneal Injection for 13 Weeks**

IV. DISCUSSION

Because soluble antimony compounds are poorly absorbed from the gastrointestinal tract and cause gastrointestinal irritation and vomiting, parenteral injections were used for treatment of schistosomiasis and leishmaniasis (AMA, 1983). There is a low therapeutic ratio of APT, and chemical treatment for parasites in humans often resulted in severe toxicity, including weight loss, myocardial insufficiency, glomerular nephritis, jaundice, erythrocytosis, and leukopenia (Venogupal and Luckey, 1978). These parasitic infections account for more than 1 million human deaths annually (AMA, 1983). Although newer drugs with fewer toxic side effects are now available, until recently APT remained the drug of choice based upon economic considerations (personal communication, E. J. Martin, FDA) and treatment efficacy for *Schistosoma japonicum* infections (Gilman *et al.,* 1990; AMA, 1983). Now, APT has been replaced by praziquantel which has fewer toxic side effects and is better tolerated by humans (K. E. Mott, WHO, Chief, Schistosomiasis Research Program, personal communication, 1990).

Comparison of the concentrations of antimony in tissues of rats given APT for 14-16 days showed about twice as much in the blood of rats given APT in the drinking water, compared to rats given i.p. injections. Since the animals in the drinking water study were given approximately 10-fold higher doses of APT, absorption of the compound appeared to be lower than after i.p. administration. There were similar concentrations of antimony present in the kidney, heart, spleen, and liver after either route of administration. The complex antimony salt could have been tightly bound or complexed to the erythrocytes, as shown for other antimony salts (Hoover, 1975). Erythrocytes have an affinity for some antimony compounds (Otto and Maren, 1950), and this may explain the higher concentrations of antimony measured in the blood compared to the remaining tissues. In the 13-week i.p. injection study, antimony continued to accumulate in proportion to dose in the spleen, liver, kidney, and heart, although the higher blood:tissue ratio of antimony was maintained.

The i.p. route of administration was selected for further studies of APT toxicity to better simulate the treatment regimen that was used in humans, and because the chemical apparently was poorly absorbed by the oral route. When antimony was administered by the oral route, the primary findings in rats and mice were decreased water consumption and weight gains; histopathological evidence of chemical toxicity was limited to mice administered the highest dose of APT. The minimal changes in the liver resembled hydropic or fatty change. Although necrosis of the forestomach in 4/10 mice was related to APT administration in the water, these lesions were focal. The remaining portions of forestomach, glandular stomach, and intestine had no gross or microscopic changes consistent with a direct irritant effect of APT. Since there were several deaths in the high dose groups of mice and rats given APT by the i.p. route, and because inflammation occurred at the site of injection in the 14-day studies, the doses and the frequency of injections were reduced in the 13-week studies. Despite reducing the injection interval to 3 times per week and to alternate sides, chronic active inflammation of the mesentery was present in treated and control animals, but these focal lesions generally were of minimal severity and did not interfere in the interpretation of the studies.

Based upon histopathology and clinical pathology, the liver was identified as the most sensitive target organ for APT toxicity. There was evidence of hepatocyte degeneration and necrosis, accompanied by dose-related elevations in activities of liver-specific serum enzymes derived from the cytosolic cell compartment. Alanine aminotransferase and sorbitol dehydrogenase serum enzyme activities exhibited dose-related elevations after 45 or 94 days of treatment. These two serum enzymes were equally useful for detecting liver toxicity, whereas lactate dehydrogenase and alkaline phosphatase enzyme activities and total bilirubin concentration in the serum were insensitive at the lower doses of APT. In rats dosed with the 24 mg/kg (high dose) of APT, there were further 3- to 10-fold elevations in these enzyme activities, and also significant elevations in serum bilirubin concentration, indicating that hepatocellular toxicity was dose-dependent. A local irritant effect of antimony is unlikely the primary cause for the necrosis, inflammation, and fibrosis along the capsular surface that was seen in most rats at the top 3 doses. In the 14-day study, APT did not produce the capsule lesions in the liver even when administered at equivalent doses and with a more frequent, daily dose schedule. The morphologic appearance and variable distribution of this capsular lesion in the 13-week study may be related to the position of the liver lobes in the abdominal cavity. Portions of liver lobes adjacent to the abdominal wall may have had greater exposure to the injected antimony solution prior to its complete absorption from the abdominal cavity. Accumulation of greater concentrations of APT may have occurred in these subcapsular hepatocytes. Further evidence of a cumulative effect for toxicity is supported by the absence of scattered foci of hepatocellular degeneration and necrosis and biliary hyperplasia in the 14-day study. These were seen in the 13-week study despite the fact that the dose schedule was reduced to 3 times per week. In mice, the liver toxicity evident in the 14-day study was not seen in the 13-week study. This may have been because of the lower doses administered and the alternate day dosing schedule in the 13-week study.

Although degeneration of the kidney was seen in only 1 of 5 male rats from the 16-day intraperitoneal study, renal toxicity was evident in 3 male rats administered the highest dose of APT in the 13-week study. The distribution of tubular cell degeneration and necrosis (outer stripe of the outer medulla) seen in this study has been attributed to the toxicity of metals or to ischemic injury to the kidney (Bulger, 1986). The presence of tubular cell regeneration in association with the degeneration in 2 rats which survived until the terminal sacrifice suggests the renal effect is more likely a result of chemical toxicity than a terminal hypoxic change in the kidney of moribund animals.

Although deaths occurred in male rats in the high-dose group in the 13-week study, there was little evidence of injury to organs, besides the liver, suspected as potential sites of APT toxicity. There was minimal evidence of kidney toxicity. There were no renal lesions in mice, and only 3 rats in the high dose group exhibited treatment-related degeneration of the kidney. Also, there was an absence of a response in a panel of 6 urinary enzymes employed as biochemical markers for renal tubular lesions in the rat studies (Zalups and Diamond, 1987). In addition, there was no evidence of toxicity to the heart based on histological examination or on measurements of LDH and CPK isoenzyme responses used to identify cardiotoxicity in the rat studies (Kaplan and Pesce, 1984); nor were there biologically significant changes in either rodent species in a panel of hematological measurements. These data indicated that because rodent hepatotoxicity occurred well before renal toxicity, monitoring the activities of liver-

specific serum enzymes during chronic studies could be a method to closely monitor the progression of APT toxicity.

The F344/N rat would serve as an acceptable animal model for any further chronic studies of APT that might be required. The i.p. route is a reasonable substitute for the i.v. route that was used in humans, and although prolonged treatment caused some mesenteric inflammation in rodents, by alternating the days of administration and the site of injection this regimen should not interfere with the interpretation of toxicity and carcinogenicity data derived from longer-term studies.

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APPENDIX A

Organ Weights in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate

Table A1 Organ Weight and Organ-Weight to Body Weight Ratios for Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate A-2

Table A2 Organ Weight and Organ-Weight to Body Weight Ratios for Mice in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate A-4

TABLE A1
Organ Weight and Organ-Weight-to-Body-Weight Ratios for Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Organ	0 mg/kg	1.5 mg/kg	3 mg/kg	6 mg/kg	12 mg /kg	24 mg/kg[b]
Male						
Necropsy body wt (g)	383 ± 7	394 ± 9	386 ± 9	387 ± 8	353 ± 7*	322 ± 9**
Brain						
Absolute	1 95 ± 0 01	2 02 ± 0 03	1 95 ± 0 03	1 99 ± 0 02	1 95 ± 0 02	1 91 ± 0 03
Relative	5 11 ± 0 08	5 14 ± 0 09	5 06 ± 0 06	5 15 ± 0 08	5 52 ± 0 07**	5 94 ± 0 14**
Heart						
Absolute	1 107 ± 0 028	1 201 ± 0 035	1 133 ± 0 040	1 153 ± 0 038	1 081 ± 0 045	0 979 ± 0 029*
Relative	2 89 ± 0 07	3 06 ± 0 08	2 93 ± 0 06	2 98 ± 0 07	3 05 ± 0 09	3 05 ± 0 05
R Kidney						
Absolute	1 26 ± 0 04	1 34 ± 0 03	1 27 ± 0 05	1 31 ± 0 03	1 26 ± 0 03	1 30 ± 0 06
Relative	3 28 ± 0 06	3 40 ± 0 05	3 27 ± 0 08	3 40 ± 0 05	3 56 ± 0 08**	4 03 ± 0 11**
Liver						
Absolute	13 55 ± 0 41	15 34 ± 0 41*	14 81 ± 0 65	15 78 ± 0 63*	14 06 ± 0 44	13 69 ± 0 56
Relative	35 4 ± 0 54	39 0 ± 0 69**	38 2 ± 0 97**	40 7 ± 1 05**	39 8 ± 0 70**	42 5 ± 0 74**
Lungs						
Absolute	2 04 ± 0 06	2 58 ± 0 16	2 23 ± 0 19	2 12 ± 0 11	2 03 ± 0 14	2 01 ± 0 10
Relative	5 33 ± 0 14	6 56 ± 0 40	5 73 ± 0 39	5 46 ± 0 22	5 80 ± 0 48	6 26 ± 0 27
Spleen						
Absolute	0 836 ± 0 017	0 898 ± 0 021	0 857 ± 0 032	0 881 ± 0 019	0 920 ± 0 08	0 733 ± 0 015
Relative	2 19 ± 0 03	2 28 ± 0 03*	2 21 ± 0 05	2 28 ± 0 04	2 60 ± 0 21**	2 29 ± 0 07*
R Testis						
Absolute	1 48 ± 0 03	1 50 ± 0 03	1 43 ± 0 04	1 56 ± 0 05	1 32 ± 0 09	1 37 ± 0 03*
Relative	3 87 ± 0 07	3 81 ± 0 07	3 70 ± 0 08	4 03 ± 0 14	3 75 ± 0 26	4 27 ± 0 15*
Thymus						
Absolute	0 383 ± 0 022	0 443 ± 0 023	0 392 ± 0 032	0 450 ± 0 039	0 352 ± 0 016	0 289 ± 0 020*
Relative	1 00 ± 0 05	1 13 ± 0 06	1 01 ± 0 08	1 16 ± 0 091	1 00 ± 0 042	0 90 ± 0 047
Female						
Necropsy body wt (g)	227 ± 3	220 ± 3	221 ± 3	216 ± 3*	224 ± 4	206 ± 3**
Brain						
Absolute	1 83 ± 0 02	1 79 ± 0 02	1 80 ± 0 02	1 76 ± 0 02*	1 81 ± 0 02	1 75 ± 0 01**
Relative	8 09 ± 0 11	8 15 ± 0 15	8 15 ± 0 11	8 19 ± 0 14	8 09 ± 0 13	8 52 ± 0 13
Heart						
Absolute	0 752 ± 0 014	0 730 ± 0 019	0 726 ± 0 018	0 719 ± 0 014	0 743 ± 0 020	0 716 ± 0 015
Relative	3 32 ± 0 05	3 32 ± 0 07	3 28 ± 0 08	3 33 ± 0 07	3 33 ± 0 13	3 47 ± 0 05
R Kidney						
Absolute	0 738 ± 0 014	0 826 ± 0 016**	0 761 ± 0 018*	0 734 ± 0 012	0 806 ± 0 017*	0 812 ± 0 012**
Relative	3 26 ± 0 07	3 76 ± 0 07**	3 44 ± 0 05**	3 40 ± 0 06*	3 60 ± 0 05**	3 94 ± 0 06**
Liver						
Absolute	6 91 ± 0 09	8 07 ± 0 17**	7 29 ± 0 18**	7 40 ± 0 10**	7 97 ± 0 17**	8 65 ± 0 14**
Relative	30 5 ± 0 38	36 7 ± 0 85**	32 9 ± 0 47**	34 3 ± 0 55**	35 6 ± 0 53**	42 0 ± 0 82**
Lungs						
Absolute	1.58 ± 0 08	1 47 ± 0 04	1 53 ± 0 05	1 58 ± 0 05	1 61 ± 0 09	1 33 ± 0 03**
Relative	6 99 ± 0 38	6 69 ± 0 22	6 92 ± 0 23	7 34 ± 0 24	7 19 ± 0 39	6 44 ± 0 09
Spleen						
Absolute	0 544 ± 0 008	0 528 ± 0 015	0 578 ± 0 014	0 564 ± 0 014	0.597 ± 0 009**	0 565 ± 0 011*
Relative	2.40 ± 0 03	2 40 ± 0 07	2.62 ± 0 07**	2 61 ± 0 07**	2.67 ± 0 05**	2 74 ± 0 04**

TABLE A1
Organ Weight and Organ-Weight-to-Body-Weight Ratios for Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a] (continued)

Organ	0 mg/kg	1.5 mg/kg	3 mg/kg	6 mg/kg	12 mg /kg	24 mg/kg[b]
Female (continued)						
Thymus						
Absolute	0 300 ± 0.012	0 284 ± 0.008	0.324 ± 0.016	0.309 ± 0.018	0 327 ± 0.017	0 252 ± 0 005*
Relative	1.33 ± 0.06	1.29 ± 0.05	1.46 ± 0.08	1.44 ± 0.09	1.46 ± 0 07	1 22 ± 0.03

* Significantly different (P≤0.05) from the control group by Dunn's or Shirley's test
** P≤0.01
[a] Organ weights are given as grams (mean ± standard error). Organ-weight-to-body weight ratios are given as mg organ weight/g body weight (mean ± standard error); n=10 for all groups except where noted
[b] n=6 for males

TABLE A2
Organ Weight and Organ-Weight-to-Body-Weight Ratios for Mice in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Organ	0 mg/kg[b]	1.5 mg/kg	3 mg/kg[b]	6 mg/kg	12 mg/kg	24 mg/kg
Male						
Necropsy body wt (g)	32 9 ± 0 7	31 2 ± 0 5	31 4 ± 1 0	33 1 ± 0 6	32 2 ± 0 9	31 3 ± 0 7
Brain						
Absolute	0 452 ± 0 004	0 453 ± 0 004	0 449 ± 0 005	0 453 ± 0 007	0 449 ± 0 008	0 456 ± 0 005
Relative	13 8 ± 0 24	14 5 ± 0 24	14 4 ± 0 44	13 7 ± 0 20	14 0 ± 0 32	14 6 ± 0 29
Heart						
Absolute	0 165 ± 0 008	0 152 ± 0 004	0 156 ± 0 006	0 150 ± 0 003	0 151 ± 0 003	0 150 ± 0 008
Relative	5 00 ± 0 20	4 87 ± 0 11	4 96 ± 0 10	4 57 ± 0 14	4 71 ± 0 08	4 78 ± 0 20
R Kidney						
Absolute	0 280 ± 0 005	0 296 ± 0 008	0 303 ± 0 010	0 262 ± 0 006	0 267 ± 0 007	0 266 ± 0 007
Relative	8 53 ± 0 20	9 47 ± 0 25	9 65 ± 0 13	7 92 ± 0 18	8 30 ± 0 20	8 50 ± 0 17
Liver						
Absolute	1 63 ± 0 06	1 53 ± 0 04	1 56 ± 0 06	1 61 ± 0 03	1 59 ± 0 05	1 62 ± 0 04
Relative	49 5 ± 1 40	49 0 ± 0 93	49 7 ± 1 29	48 7 ± 0 53	49 2 ± 1 02	51 9 ± 0 60
Lungs						
Absolute	0 327 ± 0 018	0 291 ± 0 14	0 260 ± 0 008*	0 355 ± 0 018	0 341 ± 0 015	0 323 ± 0 012
Relative	9 95 ± 0 51	9 33 ± 0 47	8 32 ± 0 29	10 74 ± 0 51	10 63 ± 0 54	10 38 ± 0 53
Spleen						
Absolute	0 070 ± 0 002	0 068 ± 0 003	0 069 ± 0 002	0 069 ± 0 001	0 073 ± 0 001	0 008 ± 0 002
Relative	2 12 ± 0 09	2 19 ± 0 07	2 20 ± 0 06	2 09 ± 0 04	2 26 ± 0 06	2 42 ± 0 07**
R Testis						
Absolute	0 118 ± 0 003	0 122 ± 0 002	0 120 ± 0 003	0 122 ± 0 002	0 121 ± 0 002	0 118 ± 0 003
Relative	3 60 ± 0 10	3 90 ± 0 08*	3 83 ± 0 10	3 69 ± 0 08	3 77 ± 0 06	3 77 ± 0 05
Thymus						
Absolute	0 050 ± 0 003	0 043 ± 0 003	0 045 ± 0 003	0 053 ± 0 005	0 056 ± 0 003	0 051 ± 0 005
Relative	1 51 ± 0 08	1 36 ± 0 10	1 45 ± 0 12	1 59 ± 0 13	1 73 ± 0 11	1 62 ± 0 16
Female						
Necropsy body wt (g)	27 2 ± 0 7	27 8 ± 0 7	27 8 ± 0 8	28 0 ± 0 9	27 4 ± 0 5	25 9 ± 0 5
Brain						
Absolute	0 455 ± 0 006	0 463 ± 0 004	0 464 ± 0 006	0 462 ± 0 005	0 467 ± 0 007	0 470 ± 0 010
Relative	16 8 ± 0 34	16 7 ± 0 33	16 8 ± 0 47	16 6 ± 0 42	17 1 ± 0 41	18 2 ± 0 48
Heart						
Absolute	0 140 ± 0 006	0 135 ± 0 003	0 134 ± 0 004	0 136 ± 0 004	0 135 ± 0 004	0 133 ± 0 004
Relative	5 13 ± 0 15	4 87 ± 0 14	4 84 ± 0 11	4 88 ± 0 17	4 93 ± 0 14	5 12 ± 0 09
R Kidney						
Absolute	0 190 ± 0 007	0 205 ± 0 008	0 207 ± 0 004	0 193 ± 0 005	0 184 ± 0 005	0 189 ± 0 004
Relative	6 98 ± 0 16	7 38 ± 0 23	7 47 ± 0 19	6 90 ± 0 19	6 71 ± 0 09	7 33 ± 0 12
Liver						
Absolute	1 34 ± 0 06	1 33 ± 0 04	1 32 ± 0 04	1 39 ± 0 04	1 41 ± 0 03	1 36 ± 0 04
Relative	49 1 ± 1 02	47 9 ± 0 78	47 6 ± 0 45	49 5 ± 0 80	51 3 ± 0 51*	52 4 ± 0 72**
Lungs						
Absolute	0 298 ± 0 012	0 263 ± 0 006	0 261 ± 0 010	0 299 ± 0 022	0 287 ± 0 009	0 298 ± 0 015
Relative	10 9 ± 0 31	9 5 ± 0 24*	9 4 ± 0 42	10 6 ± 0 50	10 5 ± 0 38	11 5 ± 0 57
Spleen						
Absolute	0 089 ± 0 005	0 084 ± 0 004	0 084 ± 0 001	0 091 ± 0 002	0 092 ± 0 004	0 087 ± 0 002
Relative	3 27 ± 0 15	3 04 ± 0 15	3 04 ± 0 07	3 28 ± 0 10	3 40 ± 0 17	3 35 ± 0 08

TABLE A2
Organ Weight and Organ-Weight-to-Body-Weight Ratios for Mice in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a] (continued)

Organ	0 mg/kg[b]	1.5 mg/kg	3 mg/kg[b]	6 mg/kg	12 mg/kg	24 mg/kg
Female (continued)						
Thymus						
Absolute	0.054 ± 0.002	0.056 ± 0.004	0 052 ± 0.003	0.057 ± 0.003	0.058 ± 0 002	0 058 ± 0 003
Relative	1.99 ± 0.05	2.01 ± 0.14	1.87 ± 0.12	2.04 ± 0.09	2.12 ± 0.09	2.24 ± 0.14

* Significantly different (P≤0.05) from the control group by Dunn's or Shirley's test
** P≤0.01
[a] Organ weights given as grams (mean ± standard error). Organ-weight-to-body weight ratios are given as mg organ weight/g body (mean ± standard error); n=10 for all groups except where noted
[b] n=9 for males

APPENDIX B

Hematology, Clinical Chemistry, and Urinalysis Data in the13-Week Intraperitoneal Studies of Antimony Potassium Tartrate

Table B1 Hematology, Clinical Chemistry, and Urinalysis Data for Special Study Rats at Day 45 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate.....B-2

Table B2 Hematology, Clinical Chemistry, and Urinalysis Data for Special Study Rats at Day 90 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate.....B-4

Table B3 Hematology and Urinalysis Data for Special Study Mice at Day 45 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate B-6

Table B4 Hematology and Urinalysis Data for Special Study Mice at Day 90 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate B-8

TABLE B1
Hematology, Clinical Chemistry, and Urinalysis Data for Rats at Day 45 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Analysis	0 mg/kg	6 mg/kg	12 mg/kg	24 mg/kg
Male				
White blood cell ($10^3/\mu L$)	8 93 ± 0 42	9 95 ± 0 33	9 83 ± 0 68	11 54 ± 1 05*[b]
Lymphocytes ($10^3/\mu L$)	7 34 ± 0 43	8 38 ± 0 30	8 04 ± 0 46	9 00 ± 0 62*[b]
Segmented neutrophils ($10^3/\mu L$)	1 42 ± 0 12	1 42 ± 0 10	1 62 ± 0 21	2 38 ± 0 61[b]
Monocytes ($10^3/\mu L$)	0 07 ± 0 03	0 07 ± 0 03	0 13 ± 0 04	0 14 ± 0 05[b]
Eosinophils ($10^3/\mu L$)	0 10 ± 0 04	0 08 ± 0 01	0 04 ± 0 02	0 02 ± 0 02[b]
Hematocrit (%)	46 2 ± 1 2	47 5 ± 0 6	45 6 ± 0 6	43 5 ± 2 6[b]
Hemoglobin (g/dL)	15 5 ± 0 4	15 8 ± 0 2	15 2 ± 0 2	14 7 ± 0 9[b]
Mean cell hemoglobin (pg)	17 4 ± 0 2	17 4 ± 0 1	17 5 ± 0 1	17 7 ± 0 2[b]
Mean cell hemoglobin concentration (g/dL)	33 6 ± 0 2	33 3 ± 0 2	33 3 ± 0 2	33 9 ± 0 3[b]
Mean cell volume (fL)	51 6 ± 0 3	52 2 ± 0 1	52 8 ± 0 3*	52 3 ± 0 6[b]
Methemoglobin (g/dL)	0 13 ± 0 01	0 15 ± 0 01	0 12 ± 0 01	0 12 ± 0 02[b]
Red blood cell ($10^6/\mu L$)	8 95 ± 0 25	9 09 ± 0 10	8 65 ± 0 12*	8 38 ± 0 57[b]
Platelets ($10^3/\mu L$)	646 8 ± 33 8	635 4 ± 20 3	549 3 ± 20 8	581 0 ± 66 1[b]
Reticulocytes ($10^6/\mu L$)	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0[b]
Alkaline phosphatase (IU/L)	636 ± 18	565 ± 20	623 ± 27	632 ± 90[b]
ALT (IU/L)	76 ± 10	88 ± 13	197 ± 36	235 ± 132[b]
Total bilirubin (mg/dL)	0 1 ± 0 0	0 1 ± 0 0	0 2 ± 0 1	0 2 ± 0 1[b]
Creatine kinase (U/L)	302 ± 36	218 ± 18[c]	379 ± 105[c]	351 ± 58[d]
LDH (IU/L)	233 ± 44	195 ± 28	274 ± 38	327 ± 79[b]
SDH (IU/L)	32 ± 5	43 ± 8	58 ± 10*	119 ± 63[d]
Creatinine phosphokinase MB (IU/L)	64 90 ± 11 41	48 70 ± 7 41	69 20 ± 14 25	54 00 ± 10 79[d]
Creatinine phosphokinase MM (IU/L)	109 8 ± 17 5	122 0 ± 41 8	192 7 ± 104[c]	162 7 ± 44 6[d]
Creatinine phosphokinase BB (IU/L)	127 6 ± 17 0	100 8 ± 10 7	156 8 ± 30 2	134 5 ± 26 8[d]
Urine gamma glutamyltransferase (mU/day)	623 ± 85[c]	725 ± 112	816 ± 208[c]	478 ± 86
Urine specific gravity	1 015 ± 0 001[c]	1 022 ± 0 002	1 020 ± 0 005[c]	1 018 ± 0 002
Urine alkaline phosphatase (IU/L)	74 ± 18[c]	108 ± 23	90 ± 28[c]	110 ± 35
Urine AST (IU/L)	10 ± 1[c]	10 ± 1	11 ± 1[c]	15 ± 3
Urine beta glucuronidase (IU/L)	16 98 ± 5 13[c]	17 10 ± 4 46	9 61 ± 3 82[c]	7 85 ± 1 75
Urine creatinine (mg/dL)	40 ± 4[c]	50 ± 5	56 ± 17[c]	43 ± 4
Urine LDH (IU/L)	17 ± 1[c]	24 ± 3	23 ± 6[c]	41 ± 13
Urine N acetyl B glucose aminidase (IU/L)	3 00 ± 0 39[c]	3 57 ± 0 69	3 29 ± 0 99[c]	2 73 ± 0 44
Urine volume (ml/16 hr)	13 4 ± 1 3[c]	11 7 ± 0 9	14 2 ± 2 5[c]	13 0 ± 1 7
Female				
White blood cell ($10^3/\mu L$)	7 99 ± 0 39	7 19 ± 0 32[c]	7 92 ± 0 64	9 29 ± 0 43
Lymphocytes ($10^3/\mu L$)	6 38 ± 0 33	6 00 ± 0 25[c]	6 75 ± 0 58	7 64 ± 0 28*
Segmented neutrophils ($10^3/\mu L$)	1 52 ± 0 11	1 10 ± 0 09[c]	1 13 ± 0 17	1 61 ± 0 19
Monocytes ($10^3/\mu L$)	0 04 ± 0 02	0 01 ± 0 01[c]	0 00 ± 0 00	0 02 ± 0 01
Eosinophils ($10^3/\mu L$)	0 06 ± 0 02	0 08 ± 0 03[c]	0 05 ± 0 02	0 03 ± 0 01
Hematocrit (%)	45 0 ± 0 5	46 1 ± 0 4[c]	44 6 ± 1 1	44 7 ± 1 2
Hemoglobin (g/dL)	14 9 ± 0 2	15 1 ± 0 2[c]	14 7 ± 0 3	14 6 ± 0 4
Mean cell hemoglobin (pg)	18 4 ± 0 1	18 4 ± 0 2[c]	18 5 ± 0 1	18 2 ± 0 1
Mean cell hemoglobin concentration (g/dL)	33 0 ± 0 1	32 7 ± 0 2[c]	33 1 ± 0 3	32 8 ± 0 2
Mean cell volume (fL)	55 7 ± 0 3	56 3 ± 0 5[c]	55 8 ± 0 3	55 4 ± 0 2
Methemoglobin (g/dL)	0 08 ± 0 02	0 05 ± 0 01[c]	0 06 ± 0 01	0 04 ± 0 02
Red blood cell (10^6 µL)	8 09 ± 0 11	8 20 ± 0 10[c]	7 97 ± 0 18	8 06 ± 0 23
Platelets ($10^3/\mu L$)	610 1 ± 13 7	602 1 ± 18 9[c]	596 8 ± 22 1	603 9 ± 12 7

TABLE B1
Hematology, Clinical Chemistry, and Urinalysis Data for Rats at Day 45 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a] (continued)

Analysis	0 mg/kg	6 mg/kg	12 mg/kg	24 mg/kg
Female (continued)				
Reticulocytes ($10^6/\mu L$)	0 2 ± 0 0	0 2 ± 0 0[c]	0 2 ± 0 0	0 1 ± 0 0**
Alkaline phosphatase (IU/L)	457 ± 21	416 ± 12	423 ± 14	434 ± 15
ALT (IU/L)	51 ± 4	38 ± 3	45 ± 3	57 ± 6
Total bilirubin (mg/dL)	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0
Creatine kinase (U/L)	330 ± 49	354 ± 51	345 ± 45	383 ± 44
LDH (IU/L)	267 ± 29	315 ± 41	288 ± 24	285 ± 43
SDH (IU/L)	36 ± 4	25 ± 3*	29 ± 1	30 ± 3
Creatinine phosphokinase MB (IU/L)	21 30 ± 2.55	21 00 ± 1 14[c]	21 89 ± 2 54[c]	22 00 ± 2 07
Creatinine phosphokinase MM (IU/L)	159 9 ± 34 9	166 4 ± 40 8	163 7 ± 40 3	203 5 ± 32 0
Creatinine phosphokinase BB (IU/L)	148 7 ± 18 0	157 8 ± 15 1	150 5 ± 15 0	157 5 ± 23 0
Urine gamma-glutamyltransferase (mU/day)	370 ± 50	270 ± 46	191 ± 24**[c]	221 ± 54*
Urine specific gravity	1 018 ± 0 002	1 017 ± 0 003	1 014 ± 0 001	1 015 ± 0 003*
Urine alkaline phosphatase (IU/L)	189 ± 66	71 ± 23	48 ± 21*	26 ±13**
Urine AST (IU/L)	13 ± 3	8 ± 1	7 ± 1	10 ± 2
Urine beta glucuronidase (IU/L)	3 66 ± 0 37	5 40 ± 0 90	3 37 ± 0 86	3 56 ± 1 15
Urine creatinine (mg/dL)	42 ± 4	37 ± 7	29 ± 3*	35 ± 8*
Urine LDH (IU/L)	17 ± 3	16 ± 3	11 ± 3	14 ± 4
Urine N-acetyl ß glucose aminidase (IU/L)	3 28 ± 0 40	2 76 ± 0 73	1 98 ± 0 35*	2 62 ± 0 77
Urine volume (ml/16 hr)	7 7 ± 0 7	8 9 ± 1 2	10 7 ± 1 1	9 4 ± 1 8

* Significantly different ($P \le 0.05$) from the control group by Dunn's or Shirley's test
** $P \le 0.01$
[a] Data given as mean ± standard error n=10 for all groups except where noted P values are vs the controls by Dunn's test (Dunn, 1964) or Shirley's test (Shirley 1977), ALT=alanine aminotransferase LDH=lactate dehydrogenase, SDH=sorbitol dehydrogenase, AST=aspartate aminotransferase
[b] n=7
[c] n=9
[d] n=6

TABLE B2
Hematology, Clinical Chemistry, and Urinalysis Data for Rats at Day 90 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Analysis	0 mg/kg	6 mg/kg	12 mg/kg	24 mg/kg[b]
Male				
n	10	10	10	9
White blood cell ($10^3/\mu L$)	7 41 ± 0 29	7 95 ± 0 25	6 72 ± 0 29	7 19 ± 0 27
Lymphocytes ($10^3/\mu L$)	6 08 ± 0 28	6 04 ± 0 26	5 27 ± 0 24	5 06 ± 0 32*
Segmented neutrophils ($10^3/\mu L$)	1 21 ± 0 08	1 84 ± 0 14**	1 41 ± 0 16	2 08 ± 0 19**
Eosinophils ($10^3/\mu L$)	0 12 ± 0 04	0 06 ± 0 02	0 03 ± 0 02	0 05 ± 0 02
Hematocrit (%)	50 6 ± 0 3	49 0 ± 0 7	48 1 ± 0 7*	50 1 ± 0 8
Hemoglobin (g/dL)	15 6 ± 0 1	14 9 ± 0 2*	14 9 ± 0 2*	15 4 ± 0 3
Mean cell hemoglobin (pg)	17 1 ± 0 1	17 0 ± 0 1	17 2 ± 0 1	17 1 ± 0 1
Mean cell hemoglobin concentration (g/dL)	30 7 ± 0 2	30 4 ± 0 1	30 9 ± 0 2	30 7 ± 0 1
Mean cell volume (fL)	55 5 ± 0 5	55 8 ± 0 3	55 7 ± 0 5	55 7 ± 0 2
Red blood cell ($10^6/\mu L$)	9 13 ± 0 09	8 80 ± 0 12	8 63 ± 0 15*	9 00 ± 0 14
Platelets ($10^3/\mu L$)	530 5 ± 10 8	591 3 ± 26 7	596 4 ± 26 9	545 9 ± 16 2
Reticulocytes ($10^6/\mu L$)	0 2 ± 0 0	0 3 ± 0 0*	0 3 ± 0 0	0 2 ± 0 0
Alkaline phosphatase (IU/L)	518 ± 14	441 ± 17**	434 ± 6**	459 ± 29**
ALT (IU/L)	93 ± 11	91 ± 8	203 ± 32**	1416 ± 339**
Total bilirubin (mg/dL)	0 0 ± 0 0	0 0 ± 0 0	0 0 ± 0 0	0 1 ± 0 0**
Creatine kinase (U/L)	237 ± 45	250 ± 23	247 ± 33	249 ± 24
LDH (IU/L)	255 ± 41	246 ± 35	317 ± 47	379 ± 52*
SDH (IU/L)	53 ± 9	51 ± 6	94 ± 10**	686 ± 148
Creatinine phosphokinase MB (IU/L)	27 70 ± 4 85	25 50 ± 3 54	27 00 ± 5 16	25 78 ± 1 85**
Creatinine phosphokinase MM (IU/L)	117 50 ± 21 81	111 50 ± 19 10	89 30 ± 12 12	100 67 ± 18 90
Creatinine phosphokinase BB (IU/L)	115 7 ± 26 6	112.6 ± 13 8	130 8 ± 20 8	123 1 ± 20 7
Urine gamma-glutamyltransferase (mU/day)	871 ± 83	748 ± 97	792 ± 62	746 ± 142
Urine specific gravity	1 025 ± 0 002	1 037 ± 0 017	1 020 ± 0 002	1 021 ± 0 002
Urine alkaline phosphatase (IU/L)	273 ± 22	574 ± 342	236 ± 27	220 ± 31
Urine AST (IU/L)	6 ± 0	16 ± 11	5 ± 0	5 ± 2*
Urine beta glucuronidase (IU/L)	17 15 ± 3 87	18 43 ± 5 13	23 48 ± 8 76	7 99 ± 2 29
Urine creatinine (mg/dL)	77 ± 7	58 ± 6	60 ± 6	64 ± 7
Urine LDH (IU/L)	30 ± 3	22 ± 3	21 ± 2*	25 ± 4
Urine N acetyl β-glucose aminidase (IU/L)	4 91 ± 0 46	4 23 ± 0 57	3 45 ± 0 37*	3 93 ± 0 34*
Urine volume (ml/16 hr)	10 7 ± 1 0	12 5 ± 1 7	10 8 ± 0 9	11 1 ± 1 4
Female				
n	9	10	9	9
White blood cell ($10^3/\mu L$)	7 82 ± 0 42	7 76 ± 0 36	7 71 ± 0 53	8 59 ± 0 47
Lymphocytes ($10^3/\mu L$)	6 12 ± 0 44	6 33 ± 0 35	6 32 ± 0 40	6 84 ± 0 38
Segmented neutrophils (10^3 μL)	1 58 ± 0 18	1 39 ± 0 09	1 23 ± 0 22	1 72 ± 0 14
Monocytes ($10^3/\mu L$)	0 01 ± 0 01	0 01 ± 0 01	0 00 ± 0 00	0 00 ± 0 00
Eosinophils ($10^3/\mu L$)	0 11 ± 0 03	0 03 ± 0 01*	0 04 ± 0 02*	0 03 ± 0 02**
Hematocrit (%)	46 7 ± 0 5	47 4 ± 0 3	47 0 ± 0 6	48 2 ± 0 5*
Hemoglobin (g/dL)	14 8 ± 0 2	14 9 ± 0 1	14 9 ± 0 2	15 2 ± 0 1
Mean cell hemoglobin (pg)	18 1 ± 0 1	18 2 ± 0 1	18 4 ± 0 1	18 2 ± 0 2
Mean cell hemoglobin concentration (g/dL)	31 7 ± 0 2	31 5 ± 0 2	31 6 ± 0 2	31 6 ± 0 2
Mean cell volume (fL)	57 2 ± 0 2	57 8 ± 0 2	58 1 ± 0 2	57 8 ± 0 4
Red blood cell ($10^6/\mu L$)	8 17 ± 0 08	8 21 ± 0 06	8 09 ± 0 11	8 35 ± 0 11
Platelets ($10^3/\mu L$)	502 9 ± 24 9c	542 5 ± 12 6	501 0 ± 16 1	567 8 ± 14 9
Reticulocytes ($10^6/\mu L$)	0 2 ± 0 0	0 2 ± 0 0	0 1 ± 0 0	0 1 ± 0 0

TABLE B2
Hematology, Clinical Chemistry, and Urinalysis Data for Rats at Day 90 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a] (continued)

Analysis	0 mg/kg	6 mg/kg	12 mg/kg	24 mg/kg[b]
Female (continued)				
n	10	10	10	10
Alkaline phosphatase (IU/L)	338 ± 14	345 ± 16	363 ± 11	356 ± 15
ALT (IU/L)	63 ± 7	48 ± 1	63 ± 4	257 ± 46**
Total bilirubin (mg/dL)	0 0 ± 0 0	0 0 ± 0 0	0 0 ± 0 0	0 1 ± 0 0**
Creatine kinase (U/L)	252 ± 51	157 ± 25	167 ± 23	170 ± 19
LDH (IU/L)	205 ± 43	104 ± 16	95 ± 11	120 ± 12
SDH (IU/L)	20 ± 2	22 ± 1	40 ± 5**	150 ± 22*
Creatinine phosphokinase-MB (IU/L)	26 30 ± 3 71	18 80 ± 1 45*	18 22 ± 1 31[b]	17 60 ± 1 33*
Creatinine phosphokinase-MM (IU/L)	125 50 ± 40 02	78 50 ± 21 41	73 40 ± 12 30	85 10 ± 16 21
Creatinine phosphokinase BB (IU/L)	100 30 ± 18 44	59 50 ± 8 10	67 60 ± 12 50	65 30 ± 5 95
Urine gamma-glutamyltransferase (mU/day)	369 ± 27	290 ± 42	373 ± 60	291 ± 44
Urine specific gravity	1 022 ± 0 002	1 025 ± 0 004	1 023 ± 0 002	1 016 ± 0 002
Urine alkaline phosphatase (IU/L)	129 ± 19	102 ± 18	96 ± 18	65 ± 10**
Urine AST (IU/L)	4 ± 0	4 ± 1	4 ± 0	5 ± 1
Urine beta-glucuronidase (IU/L)	4 22 ± 0 86	4 07 ± 0 43	7 73 ± 1 61	2 99 ± 0 46
Urine creatinine (mg/dL)	55 ± 6	58 ± 9	55 ± 6	42 ± 6
Urine LDH (IU/L)	16 ± 1	19 ± 3	18 ± 1	16 ± 3
Urine N-acetyl B-glucose aminidase (IU/L)	4 55 ± 0 55	5 00 ± 0 78	4 13 ± 0 52	2 59 ± 0 45*
Urine volume (ml/16 hr)	6 7 ± 0 9	7 1 ± 1 1	6 9 ± 1 1	9 8 ± 1 6

* Significantly different (P≤0 05) from the control group by Dunn's or Shirley's test
** P≤0 01
[a] Data given as mean ± standard error n=10 for all groups except where noted P values are vs the controls by Dunn s test (Dunn, 1964) or Shirley s test (Shirley 1977), ALT=alanine aminotransferase, LDH=lactate dehydrogenase, SDH=sorbitol dehydrogenase, AST=aspartate aminotransferase
[b] n=9
[c] n=8

TABLE B3
Hematology and Urinalysis Data for Mice at Day 45 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Analysis	0 mg/kg	6 mg/kg	12 mg/kg	24 mg/kg
Male				
White blood cell ($10^3/\mu L$)	5 71 ± 0 39	5 56 ± 0 16	5 08 ± 0 34[b]	4 68 ± 0 31*
Lymphocytes ($10^3/\mu L$)	4 66 ± 0 39	4 54 ± 0 11	4 18 ± 0 34[b]	3 63 ± 0 37*
Segmented neutrophils ($10^3/\mu L$)	0 89 ± 0 14	0 81 ± 0 12	0 71 ± 0 10[b]	0 96 ± 0 22
Eosinophils ($10^3/\mu L$)	0 16 ± 0 04	0 21 ± 0 05	0 15 ± 0 04[b]	0 08 ± 0 02
Hematocrit (%)	50 2 ± 0 6	50 4 ± 0 6	49 5 ± 0 7[b]	46 5 ± 1 0**
Hemoglobin (g/dL)	15 9 ± 0 2	15 9 ± 0 2	15 6 ± 0 2[b]	14 8 ± 0 3**
Mean cell hemoglobin (pg)	15 5 ± 0 1	15 6 ± 0 1	15 8 ± 0 1[b]	16 2 ± 0 1**
Mean cell hemoglobin concentration (g/dL)	31 7 ± 0 2	31 6 ± 0 2	31 5 ± 0 3[b]	32 0 ± 0 3
Mean cell volume (fL)	48 9 ± 0 2	49 3 ± 0 3	50 1 ± 0 6[b]	50 7 ± 0 5**
Methemoglobin (g/dL)	0 07 ± 0 02	0 07 ± 0 02	0 08 ± 0 02[b]	0 05 ± 0 01
Red blood cell ($10^6/\mu L$)	10 23 ± 0 12	10 20 ± 0 14	9 90 ± 0 19[b]	9 16 ± 0 20**
Platelets ($10^3/\mu L$)	668 0 ± 40 8	654 1 ± 31 5	698 3 ± 35 4[b]	802 7 ± 40 7*
Reticulocytes ($10^6/\mu L$)	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0[b]	0 1 ± 0 0
Urine gamma-glutamyltransferase (mU/day)	182 ± 44	100 ± 11	111 ± 37*	64 ± 10**
Urine specific gravity	1 032 ± 0 002	1 028 ± 0 001	1 030 ± 0 004	1 025 ± 0 003*
Urine alkaline phosphatase (IU/L)	290 ± 146	238 ± 152	177 ± 135*	154 ± 78
Urine AST (IU/L)	8 ± 3	7 ± 3	4 ± 1	7 ± 3
Urine beta-glucuronidase (IU/L)	6 39 ± 0 39	5 47 ± 0 31	5 04 ± 0 61*	4 28 ± 0 46**
Urine creatinine (mg/dL)	26 ± 2	23 ± 1	22 ± 4	19 ± 3*
Urine LDH (IU/L)	26 ± 2	21 ± 2[b]	20 ± 3*	18 ± 2*
Urine N-acetyl B-glucose aminidase (IU/L)	41 68 ± 3 42	34 38 ± 2 52[b]	27 38 ± 3 25**	25 27 ± 3 91**
Urine volume (ml/16 hr)	1 0 ± 0 1	1 2 ± 0 2	1 5 ± 0 2	1 9 ± 0 4
Females				
White blood cell ($10^3/\mu L$)	6 29 ± 0 31	6 15 ± 0 45	5 84 ± 0 31	4 94 ± 0 30*
Lymphocytes ($10^3/\mu L$)	5 34 ± 0 36	5 25 ± 0 38	5 07 ± 0 29	4 12 ± 0 24*
Segmented neutrophils ($10^3/\mu L$)	0 77 ± 0 12	0 78 ± 0 14	0 65 ± 0 11	0 68 ± 0 11
Eosinophils ($10^3/\mu L$)	0 17 ± 0 04	0 12 ± 0 03	0 12 ± 0 03	0 14 ± 0 03
Hematocrit (%)	50 3 ± 0 8	49 8 ± 0 9	48 9 ± 0 7	47 2 ± 0 6**
Hemoglobin (g/dL)	16 4 ± 0 2	16 2 ± 0 2	15 8 ± 0 2*	15 4 ± 0 1**
Mean cell hemoglobin (pg)	15 8 ± 0 1	15 9 ± 0 1	15 8 ± 0 2	16 4 ± 0 1**
Mean cell hemoglobin concentration (g/dL)	32 7 ± 0 3	32 6 ± 0 3	32 4 ± 0 3	32 7 ± 0 3
Mean cell volume (fL)	50 3 ± 0 8	49 8 ± 0 9	48 9 ± 0 5	50 3 ± 0 5
Methemoglobin (g/dL)	0 05 ± 0 01	0 07 ± 0 01	0 08 ± 0 02	0 07 ± 0 02
Red blood cell ($10^6/\mu L$)	10 44 ± 0 16	10 16 ± 0 15	10 01 ± 0 13	9 38 ± 0 08**
Platelets ($10^3/\mu L$)	581 9 ± 31 4	602 2 ± 32 4	592 9 ± 27 7	635 9 ± 32 5
Reticulocytes ($10^6/\mu L$)	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0	0 1 ± 0 0
Urine gamma-glutamyltransferase (mU/day)	124 ± 16	107 ± 19	160 ± 18	81 ± 15
Urine specific gravity	1 020 ± 0 002	1 019 ± 0 003	1 016 ± 0 001	1 016 ± 0 003
Urine alkaline phosphatase (IU/L)	361 ± 185	615 ± 367	249 ± 113	360 ± 191
Urine AST (IU/L)	10 ± 5	5 ± 1	4 ± 1	8 ± 4
Urine beta-glucuronidase (IU/L)	2 01 ± 0 32	2 20 ± 0 38	2 12 ± 0 15	1 92 ± 0 24
Urine creatinine (mg/dL)	16 ± 2	16 ± 3	12 ± 1	13 ± 3
Urine LDH (IU/L)	15 ± 2	13 ± 2	13 ± 1	12 ± 2
Urine N-acetyl B-glucose aminidase (IU/L)	5 86 ± 1 09	5 76 ± 1 34	3 79 ± 0 47	3 89 ± 0 87
Urine volume (ml/16 hr)	1 7 ± 0 5	2 5 ± 0 6	2 6 ± 0 4	2.7 ± 0 5

TABLE B3
Hematology and Urinalysis Data for Mice at Day 45 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a] (continued)

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* Significantly different (P≤0.05) from the control group by Dunn's or Shirley's test
** P≤0.01
[a] Data given as mean ± standard error; n=10 for all groups except where noted. P values are vs. the controls by Dunn's test (Dunn, 1964) or Shirley's test (Shirley, 1977); AST=aspartate aminotransferase; LDH=lactate dehydrogenase
[b] n=9

TABLE B4
Hematology and Urinalysis Data for Mice at Day 90 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Analysis	0 mg/kg	6 mg/kg	12 mg/kg	24 mg/kg
Male				
White blood cell ($10^3/\mu L$)	6.55 ± 0.27[b]	5.87 ± 0.33	5.10 ± 0.21**	5.60 ± 0.50**
Lymphocytes ($10^3/\mu L$)	5.21 ± 0.21[b]	4.48 ± 0.37	4.14 ± 0.18*	4.84 ± 0.39
Segmented neutrophils ($10^3/\mu L$)	1.22 ± 0.13[b]	1.25 ± 0.37	0.80 ± 0.08*	0.61 ± 0.10**
Eosinophils ($10^3/\mu L$)	0.12 ± 0.06[b]	0.13 ± 0.06	0.16 ± 0.04	0.15 ± 0.05
Hematocrit (%)	59.2 ± 1.1[b]	58.5 ± 1.4	56.8 ± 1.0	56.7 ± 0.9
Hemoglobin (g/dL)	16.7 ± 0.3[b]	16.5 ± 0.5	15.9 ± 0.2*	16.1 ± 0.2*
Mean cell hemoglobin (pg)	15.3 ± 0.1[b]	15.7 ± 0.1	15.8 ± 0.1**	16.1 ± 0.1**
Mean cell hemoglobin concentration (g/dL)	28.2 ± 0.3[b]	28.3 ± 0.3	28.0 ± 0.2	28.3 ± 0.3
Mean cell volume (fL)	54.1 ± 0.6[b]	55.4 ± 0.4	56.6 ± 0.3**	57.0 ± 0.3**
Methemoglobin (g/dL)	0.02 ± 0.01[b]	0.06 ± 0.02	0.02 ± 0.01	0.06 ± 0.01
Red blood cell ($10^6/\mu L$)	10.93 ± 0.16[b]	10.58 ± 0.32	10.07 ± 0.16**	9.96 ± 0.13**
Platelets ($10^3/\mu L$)	668.0 ± 30.1[c]	670.3 ± 47.3	682.3 ± 31.0	681.0 ± 50.5
Reticulocytes ($10^6/\mu L$)	0.2 ± 0.0[c]	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.0
Urine gamma-glutamyltransferase (mU/day)	144 ± 29[c]	179 ± 40[b]	224 ± 44	136 ± 20
Urine specific gravity	1.027 ± 0.004[c]	1.033 ± 0.003	1 032 ± 0.002	1.025 ± 0.001
Urine alkaline phosphatase (IU/L)	240 ± 201[c]	296 ± 244	228 ± 99	81 ± 45
Urine AST (IU/L)	3 ± 1[c]	5 ± 3	16 ± 9	2 ± 1
Urine beta-glucuronidase (IU/L)	5.27 ± 0.77[c]	7 19 ± 0.63	6.94 ± 0.38	4.59 ± 0.35
Urine creatinine (mg/dL)	23 ± 3[c]	29 ± 4	27 ± 2	20 ± 1
Urine LDH (IU/L)	23 ± 5[c]	25 ± 5	26 ± 2	16 ± 1
Urine N-acetyl B-glucose aminidase (IU/L)	39.89 ± 4.67[c]	47.84 ± 4.53	48.62 ± 2.15	34.16 ± 2.70
Urine volume (ml/16 hr)	1.1 ± 0.2[b]	1.0 ± 0.2	1.3 ± 0.1	1.8 ± 0.3*
Females				
White blood cell ($10^3/\mu L$)	5.04 ± 0.42[d]	5.72 ± 0.33	5.52 ± 0.24[d]	4.87 ± 0.36
Lymphocytes ($10^3/\mu L$)	4 22 ± 0.34[d]	4 83 ± 0.28	4.72 ± 0.19[d]	4.04 ± 0.28
Segmented neutrophils ($10^3/\mu L$)	0.71 ± 0.10[d]	0.75 ± 0.09	0.71 ± 0.07[d]	0.75 ± 0.13
Eosinophils ($10^3/\mu L$)	0.11 ± 0.02[d]	0.14 ± 0.03	0.09 ± 0.04[d]	0.08 ± 0.01
Hematocrit (%)	56.3 ± 0.8[d]	55.8 ± 0.8	55.1 ± 0.9[d]	54.0 ± 1.4
Hemoglobin (g/dL)	16.7 ± 0.2[d]	16.6 ± 0.2	16.4 ± 0.3[d]	16.1 ± 0.4
Mean cell hemoglobin (pg)	15.7 ± 0.1[d]	15.8 ± 0.1	16.1 ± 0.1**[d]	16.4 ± 0.1**
Mean cell hemoglobin concentration (g/dL)	29.7 ± 0.2[d]	29.7 ± 0.3	29.8 ± 0.2[d]	29.9 ± 0.1
Mean cell volume (fL)	52.7 ± 0.5[d]	53.2 ± 0.4	54.3 ± 0.4**[d]	55.1 ± 0.6**
Methemoglobin (g/dL)	0.04 ± 0.02[d]	0.02 ± 0.01	0.01 ± 0.01[d]	0.03 ± 0.01
Red blood cell ($10^6/\mu L$)	10.65 ± 0.08[d]	10.48 ± 0.14	10.17 ± 0.18*[d]	9.82 ± 0.30**
Platelets ($10^3/\mu L$)	546.6 ± 45.1[d]	548.2 ± 44.1	535.6 ± 33.7[d]	569.9 ± 45.9
Reticulocytes ($10^6/\mu L$)	0.2 ± 0.0[d]	0.2 ± 0.0	0.2 ± 0.0[d]	0.2 ± 0.0
Urine gamma-glutamyltransferase (mU/day)	239 ± 56	231 ± 55	138 ± 15[d]	195 ± 32[b]
Urine specific gravity	1.022 ± 0.003	1.024 ± 0.003	1.018 ± 0.002[d]	1.020 ± 0.002[b]
Urine alkaline phosphatase (IU/L)	141 ± 39	570 ± 287	315 ± 90[d]	1687 ± 1445[b]
Urine AST (IU/L)	5 ± 1	12 ± 5	18 ± 11[d]	36 ± 27[b]
Urine beta-glucuronidase (IU/L)	2.40 ± 0.27	2.66 ± 0.46	2.32 ± 0.32[d]	2.54 ± 0.37[b]
Urine creatinine (mg/dL)	21 ± 3	22 ± 3	17 ± 3[d]	18 ± 2[b]
Urine LDH (IU/L)	14 ± 2	22 ± 5	14 ± 2[d]	15 ± 2[b]
Urine N-acetyl B-glucose aminidase (IU/L)	8.87 ± 1.04	9.84 ± 1.53	7.52 ± 1.14[d]	8.88 ± 1.39[b]
Urine volume (ml/16 hr)	2.0 ± 0.5	2.0 ± 0.5	2.8 ± 0.4[d]	2.4 ± 0.5[b]

TABLE B4
Hematology and Urinalysis Data for Mice at Day 90 in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a] (continued)

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* Significantly different ($P \leq 0.05$) from the control group by Dunn's or Shirley's test
** $P \leq 0.01$
[a] Data given as mean ± standard error; n=9 for all groups except where noted. P values are vs. the controls by Dunn's test (Dunn, 1964) or Shirley's test (Shirley, 1977); LDH=lactate dehydrogenase; AST=aspartate aminotransferase
[b] n=8
[c] n=7
[d] n=10

APPENDIX C

Results of Reproductive Analyses in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate

Table C1 Summary of Reproductive Tissue Evaluations in Male Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate C-2

Table C2 Summary of Estrous Cycle Characterization in Female Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate C-2

Table C3 Summary of Reproductive Tissue Evaluationsin Male Mice in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate C-3

Table C4 Estrous Cycle Characterization in Female Mice in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate C-3

Table C1
Summary of Reproductive Tissue Evaluations in Male Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Study Parameters	Body and Reproductive Tissue Weights and Spermatozoal Data for Control and Dosed Groups			
	0 ppm	3 mg/kg	6 mg/kg	12 mg/kg
Weights (g)				
Necropsy body weight	383 ± 7	386 ± 9	387 ± 8	353 ± 7*
L testicle	1 54 ± 0 02	1 55 ± 0 05	1 63 ± 0 02	1 46 ± 0 05
L epididymis	0 474 ± 0 008	0 454 ± 0 010	0 486 ± 0 007	0 442 ± 0 014
L epididymal tail	0 155 ± 0 004	0 152 ± 0 005	0 157 ± 0 006	0 142 ± 0 005
Spermatozoal measurements				
% motility	70 61 ± 2 35	66 22 ± 0 85	68 69 ± 1 73	72 19 ± 3 30
Concentration (10^6/g)	511 7 ± 27 0	479 0 ± 14 9	509 7 ± 26 8	476 9 ± 36 0
Spermatids (10^4/ml solution)	59 88 ± 2 97	63 39 ± 3 00	64 47 ± 2 97	59 09 ± 2 36[b]
Spermatid heads/testis(10^7)	11 98 ± 0 59	12 68 ± 0 60	12.90 ± 0 59	11 82 ± 0 47[b]
Spermatid heads/gram of testis(10^7)	7 80 ± 0 45	8 13 ± 0 26	7 93 ± 0 36	7 83 ± 0 31[b]

* Significantly different (P≤0 05) from the control group by Dunn's or Shirley's test
[a] Data presented as mean ± standard error n=10 for all groups except where noted
[b] n=9

Table C2
Summary of Estrous Cycle Characterization in Female Rats in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate

Study Parameters	Number Animals	Vaginal Cytology Data for Control and Dosed Groups			
		0 mg/kg	3 mg/kg	6 mg/kg	12 mg/kg
Necropsy body weight (g)[a]	10	227 ± 3	221 ± 3	216 ± 3*	224 ± 4
Estrous cycle length (days)[a]	10	4 70 ± 0 20	4 80 ± 0 17	4 65 ± 0 17	4 94 ± 0 18[b]
Estrous stages as % of cycle					
% diestrus	10	35 0	31 7	33 3	38 3
% proestrus	10	15 0	16 7	13 2	16 7
% estrus	10	29 2	30 0	24 6	20 8
% metestrus	10	15 0	14 2	13 2	13 3
%uncertain diagnosis	10	5 8	7 5	15 8	10 8

* Significantly different from the control group (P<0 05) by Dunn's of Shirley's test
[a] Data presented as mean ± standard error
[b] For 1/10 animals at this dose, estrus cycle length exceeded 12 days and the data was not included in the mean

Table C3
Summary of Reproductive Tissue Evaluations in Male Mice
in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Study Parameters	Body and Reproductive Tissue Weights and Spermatozoal Data for Control and Dosed Groups			
	0 ppm	6 mg/kg	12 mg/kg	24 mg/kg
Male				
Weights (g)				
Necropsy body weight	32 9 ± 0 7	33 1 ± 0 6	32 2 ± 0 9	31 3 ± 0 7
L. testicle	0 121 ± 0 003[b]	0 115 ± 0 002[b]	0 099 ± 0 010*	0 113 ± 0 002*
L. epididymis	0 048 ± 0 001[b]	0 045 ± 0 002	0 045 ± 0 002	0 047 ± 0 003
L. epididymal tail	0 014 ± 0 001[b]	0 015 ± 0 001	0 015 ± 0 001	0 016 ± 0 001
Spermatozoal measurements				
% motility	70 82 ± 2 03[b]	68 92 ± 2 26	70 19 ± 2 30[b]	70 01 ± 2 60[b]
Concentration (10^6/g)	453 0 ± 76 1[b]	449 0 ± 102	423 9 ± 68 7	368 4 ± 68 5

* Significantly different from the control group (P<0 05) by Dunn s or Shirley's test
[a] Data presented as mean ± standard error; n=10 for all groups except where noted
[b] n=9

Table C4
Estrous Cycle Characterization in Female Mice in the 13-Week Intraperitoneal Studies of Antimony Potassium Tartrate[a]

Study Parameters	Body Weights and Estrous Cycle Lengths for Dosed and Control Groups			
	0 ppm	6 mg/kg	12 mg/kg	24 mg/kg
Female				
Necropsy body weight (g)	27 2 ± 0 7	28 0 ± 0 9	27 4 ± 0 5	25 9 ± 0 5
Estrous cycle length (days)	4 11 ± 0 11	4 13 ± 0 13[b]	4 10 ± 0 10[c]	4 22 ± 0 15

* Significantly different from the control group (P<0 05) by Dunn s or Shirley s test
[a] Data presented as mean ± standard error; n=9 for all groups except where noted
[b] n=8
[c] n=10

APPENDIX D

Results of Mutagenesis Analyses of Antimony Potassium Tartrate

Table D1 Mutagenicity of Antimony Potassium Tartrate in Salmonella Typhimurium..... D-2

Table D1

Mutagenicity of Antimony Potassium Tartrate in *Salmonella Typhimurium* [a]

Strain	Dose (µg/plate)	Revertants/plate[b] (-) S9	Hamster S9 10%	Hamster S9 30%	Rat S9 10%	Rat S9 30%
TA100	0	148 ± 9.3	173 ± 5.6	160 ± 9.3	159 ± 13.4	122 ± 10.7
	10	156 ± 10.6	139 ± 1.8		163 ± 10.9	
	33	144 ± 5.1	151 ± 11.8		145 ± 10.6	
	100	136 ± 11.0	167 ± 6.3	126 ± 7.0	143 ± 7.9	117 ± 13.7
	333	126 ± 12.3	163 ± 11.5	149 ± 4.8	114 ± 5.0	145 ± 14.8
	1,000	101 ± 6.1	155 ± 5.5	125 ± 7.1	103 ± 13.9	161 ± 1.7
	3,333			94 ± 4.0		90 ± 12.4
	10,000			65 ± 11.1		39 ± 6.6
Trial Summary		Negative	Negative	Negative	Negative	Negative
Positive control[c]		316 ± 3.5	657 ± 5.1	399 ± 4.9	599 ± 23.1	447 ± 17.5
TA1535	0	13 ± 1.5	8 ± 1.2	12 ± 2.3	16 ± 3.5	11 ± 4.4
	10	15 ± 3.1	10 ± 1.7		8 ± 1.9	
	33	9 ± 0.0	6 ± 0.3		9 ± 1.9	
	100	13 ± 0.3	10 ± 1.7	12 ± 2.4	11 ± 0.9	18 ± 5.0
	333	11 ± 0.6	11 ± 1.0	12 ± 3.5	8 ± 1.5	16 ± 2.4
	1,000	10 ± 2.0	9 ± 0.3	9 ± 3.0	10 ± 2.8	15 ± 1.7
	3,333			5 ± 0.6		4 ± 0.7
	10,000			4 ± 2.3		2 ± 0.5 [d]
Trial Summary		Negative	Negative	Negative	Negative	Negative
Positive control		273 ± 10.7	150 ± 8.4	320 ± 41.3	121 ± 10.5	88 ± 6.2
TA97	0	173 ± 15.3	155 ± 5.1	177 ± 11.5	182 ± 11.5	218 ± 7.7
	10	194 ± 10.5	163 ± 6.5	187 ± 6.4	194 ± 10.5	225 ± 0.3
	33	184 ± 9.8	163 ± 12.1	190 ± 5.4	184 ± 9.8	219 ± 4.8
	100	177 ± 10.2	162 ± 5.8	173 ± 1.9	177 ± 10.2	208 ± 6.1
	333	161 ± 9.0	163 ± 8.5	131 ± 10.2	161 ± 9.0	202 ± 12.2
	1,000	137 ± 11.3	136 ± 21.0	116 ± 4.3	137 ± 11.3	135 ± 10.4
Trial Summary		Negative	Negative	Negative	Negative	Negative
Positive control		403 ± 11.1	463 ± 15.8	451 ± 19.6	449 ± 10.1	417 ± 18.2
TA98	0	21 ± 3.7	27 ± 2.8	34 ± 0.9	33 ± 3.8	32 ± 3.2
	10	25 ± 2.2	32 ± 2.9		30 ± 6.5	
	33	16 ± 2.0	30 ± 1.8		28 ± 4.6	
	100	21 ± 1.5	27 ± 5.1	28 ± 0.7	22 ± 2.2	25 ± 2.6
	333	19 ± 3.3	26 ± 2.8	35 ± 4.8	27 ± 2.2	27 ± 0.6
	1,000	23 ± 2.7	23 ± 3.3	28 ± 1.2	29 ± 2.2	33 ± 3.7
	3,333			23 ± 2.6		25 ± 1.7
	10,000			9 ± 3.5[d]		3 ± 2.3[d]
Trial Summary		Negative	Negative	Negative	Negative	Negative
Positive control		369 ± 25.1	385 ± 39.0	147 ± 9.3	279 ± 4.9	83 ± 7.3

[a] Study performed at SRI, International. The detailed protocol is presented in Zeiger et al. (1988). Cells and study compound or solvent (distilled water) were incubated in the absence of exogenous metabolic activation (-S9) or with Aroclor 1254-induced S9 from male Syrian hamster liver or male Sprague Dawley rat liver. High dose was limited by toxicity, but did not exceed 10 mg/plate; 0 µg/plate dose is the solvent control.

[b] Revertants are presented as mean ± the standard error from 3 plates.

[c] Positive control; 2-aminoanthracene was used on all strains in the presence of S9. In the absence of metabolic activation, 4-nitro-o-phenylenediamine was tested on TA98, sodium azide was tested on TA100 and TA1535, and 9-aminoacridine was tested on TA97.

[d] Slight toxicity.