



FDCN: 88420004582

8EHQ-1191-5936

UNION CARBIDE CORPORATION 39 OLD RIDGEBURY ROAD, DANBURY, CT 06817-0001



8EHQ-92-5936
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Attn: 8(e) Coordinator

RE: Union Carbide Corporation's TSCA §8(e) Submission of July 21, 1992 (et. seq.) Concerning Triethylene Glycol (CASRN 112-27-6) [8EHQ-0792-5936]

Dear Sir or Madam:

As a follow-up to the above-noted submissions concerning triethylene glycol (CASRN 112-27-6), Union Carbide Corporation ("Union Carbide") herewith submits the following report.

"Triethylene Glycol: Nine-Day Aerosol Inhalation (Nose-Only Exposure) Toxicity Study in Rats," Bushy Run Research Center, BRRC Report 93U1293, October 26, 1994 (312 pgs.). [NOTE: Only the first 20 pages of this report is enclosed; the remainder is available on written request.]

In the attached report the term "Confidential" may appear. This precautionary statement was for internal use at the time of issuance of this report. Confidentiality is hereby waived for purposes of the needs of the Agency in assessing health and safety information. The Agency is advised, however, that the publication rights to the contained information are the property of Union Carbide.

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UCC - Submission 7/21/92

Please contact the undersigned with questions, if any, at 203/794-5230.

Very truly yours,



William C. Kuryla, Ph.D.
Associate Director
Product Safety

Attachment

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STUDY TITLE

Triethylene Glycol: Nine-Day Aerosol Inhalation (Nose-Only Exposure)
Toxicity Study in Rats

TEST SUBSTANCE

Triethylene Glycol

Contains No CBI

DATA REQUIREMENT

Not Applicable

AUTHORS

J. C. Norris and W. J. Kintigh

STUDY COMPLETION DATE

October 26, 1994

PERFORMING LABORATORY

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LABORATORY PROJECT ID

93U1293

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**Triethylene Glycol: Nine-Day Aerosol Inhalation (Nose-Only Exposure)
Toxicity Study in Rats**

CONFIDENTIALITY STATEMENT

This report is Union Carbide Corporation Business Confidential and is not to be released outside of the Corporation without the written consent of the Sponsor.

**Triethylene Glycol: Nine-Day Aerosol Inhalation (Nose-Only Exposure)
Toxicity Study in Rats**

COMPLIANCE WITH GOOD LABORATORY PRACTICE STANDARDS

The portions of this study conducted by BRRC meet the requirements of the following Good Laboratory Practice Standards: Toxic Substances Control Act (TSCA), 40 CFR Part 792; Organisation for Economic Co-operation and Development (OECD), C(81)30(Final), with the following exception:

1. The prestudy GLP analysis was not performed prior to the beginning of exposures.

This exception is not expected to compromise the integrity of the results and conclusion of the study.

Study Director:


James C. Norris, Ph.D. 10/26/94
Date

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**Triethylene Glycol: Nine-Day Aerosol Inhalation (Nose-Only Exposure)
Toxicity Study in Rats**

SUMMARY

Three groups, each consisting of 10 CD¹ rats/sex, were exposed, nose-only, for 6 hours/day for 9 exposures during an 11-day period to an aerosol of triethylene glycol (TEG; CAS No. 112-27-6) at target concentrations of 100, 500, and 1000 mg/m³. A control group with the same number of rats/sex was exposed to filtered air alone. An additional 5 rats/sex were assigned to the 0 and 1000 mg/m³ group for a 4-week recovery period following the exposure regimen. Monitors for toxic effects included clinical observations, ophthalmic examinations, body and organ weights, food and water consumption, hematologic and serum clinical chemistry evaluations, protein fractions, urinalysis, urine chemistry, and necropsy and microscopic evaluations.

Mean chamber gravimetric concentrations (\pm SD) were 102 (\pm 8.2), 517 (\pm 37.9), and 1036 (\pm 27.2) mg/m³ for the target concentrations of 100, 500, and 1000 mg/m³, respectively. The mean nominal concentrations (\pm SD) were 132 (\pm 37.1), 454 (\pm 95.0), and 968 (\pm 87.8) mg/m³ for the target concentrations of 100, 500, and 1000 mg/m³, respectively. The mean analytical/nominal (A/N) concentration ratios were 0.84, 1.19, and 1.08 for TEG target concentrations of 100, 500, and 1000 mg/m³, respectively. The mean mass median aerodynamic diameters (MMAD) were 1.2, 1.4, and 1.3 microns for the 100, 500, and 1000 mg/m³ groups, respectively, with daily geometric standard deviations (G_d) ranging from 1.25 to 1.39 for all groups. The mean chamber temperature range for all exposures was 19.5 to 20.8°C.

No exposure-related clinical signs were observed in the male or female rats.

No changes in body weights for the males or females or body weight gains for the males in any TEG exposure groups were noted. A nonstatistically significant decrease in the female body weight gains of the 500 and 1000 mg/m³ groups was observed in the intervals Days 8 to 9 and 9 to 12. No changes in food and water consumption measurements for the males and females in any TEG exposure groups were noted. There were no ophthalmic lesions found in this study which were attributed to TEG exposure. No clinical pathology findings were related to the TEG exposures.

No exposure-related organ weight changes in the males or females were observed. No gross or microscopic lesions in any of the TEG-exposed animals were related to TEG exposure.

Based on the small decreased body weight gains for the females in the 500 and 1000 mg/m³ group, a no-observed-effect level (NOEL) was determined to be 100 mg/m³ for a 9-day repeated nose-only exposure to a TEG aerosol in rats.

OBJECTIVE

The objective of this study was to determine and evaluate the toxic and other effects in CD⁰ rats which may occur from a 9-day repeated nose-only aerosol inhalation exposure to triethylene glycol (TEG).

BACKGROUND INFORMATION

An acute aerosol inhalation (whole body) study was conducted in Sprague Dawley[®] rats (BRRC Report 53-53). Groups of 5 male and 5 female rats were exposed for 4 hours to an aerosol of TEG at average concentrations of 2.6, 3.9, 5.0, and 6.7 mg/l. For exposure groups \leq 3.9 mg/l, the MMAD ranged from 4.4 to 4.9 microns with σ_g ranging from 1.6 to 2.8. For exposure groups \geq 5.0 mg/l, the MMAD ranged from 5.3 to 9.3 microns with σ_g ranging from 2.7 to 3.8. All females in the 5.0 mg/l group died on exposure days 2 and 3. Following exposure these females displayed wet (oily) fur, periorcular and perinasal encrustation, a bright red discoloration of the eyes, ears, and feet, and an absence of tail and toe pinch reflexes. Gross lesions observed in these animals were periorcular and perinasal redness and/or encrustation, ulceration of the nares and tongue, dry shrunken eyes, and discoloration of the lungs, stomach, thymus, and pituitary. A repeat 5.0 mg/l group of females were exposed with no deaths prior to sacrifice. There were no deaths in any other group of females and no males at any exposure group died. Clinical signs were observed for all exposure groups on the day of exposure and for the first 2 to 5 postexposure days were wet (oily) fur, perinasal and periorcular encrustation, and discolored (brown) and unkempt fur. Additional signs observed for the 5.0 and 6.7 mg/l groups included a bright red discoloration of the eyes, ears, and feet, blepharospasm, audible respiration, decreased motor activity, and an absence of toe and tail pinch reflexes. For the 3.9 and 5.0 mg/l groups, decreases in the body weight gain occurred during the first postexposure week, but recovery occurred during the second week. No evidence of either lung or kidney injury was seen by light microscopic evaluation. Additionally, a limited repeat acute inhalation (whole body) study was conducted in Sprague Dawley[®] rats (BRRC Report 53-139). In this study, 5 male and 5 female rats were exposed for 4 hours to 5.2 mg/l of TEG. The MMAD was approximately 5.5 microns with a σ_g of 2.73. No mortality occurred during or following the exposure. Clinical signs were observed on the day of exposure and included periorcular wetness, blepharospasm, wet (oily) fur, and an absence of toe and tail pinch reflexes. Unkempt fur was the only sign observed for both sexes during the postexposure period. Mean body weight gains were observed for both sexes on postexposure Days 7 and 14. No macroscopic lesions were observed in the animals sacrificed at the end of the 14-day postexposure period. The results of this study indicated that the LC₅₀ value for TEG aerosol in Sprague Dawley[®] rats was greater than 5.2 mg/l.

A previous 9-day inhalation (whole body) study was conducted in Sprague Dawley[®] rats (BRRC Report 91U0027). In this study, 3 groups, each containing 10 rats/sex, were exposed to an aerosol of TEG at target concentrations of 500, 2000, and 5000 mg/m³ for 6 hours/day for 9 exposures. An additional 5 animals/sex were added to the high exposure and control groups for a planned recovery period. A control group was exposed to filtered air only. For the target concentrations of 500, 2000, and 5000 mg/m³, mean analytical exposure concentrations (\pm SD) were determined to be 494 (\pm 14.2), 2011 (\pm 94.4) and 4824 (\pm 182.9) mg/m³, respectively. A mean MMAD for all 3 exposure groups was

0.013

2.48 microns with a mean geometric standard deviation (σ_g) of 1.6. Determinants of toxic effects were clinical observations, ophthalmic examinations, body and organ weights, hematologic and serum clinical chemistry evaluations, urinalysis, and macroscopic and microscopic evaluations.

For all TEG-exposed rats, exposure-related clinical observations occurred. At 4824 mg/m^3 , clinical observations included ataxia; prostration, unkempt fur, labored respiration (males only), ocular discharge, swollen periocular tissue, perinasal and pericocular encrustation, and blepharospasm in both sexes unless noted otherwise. At 494 and 2011 mg/m^3 , there were swollen periocular tissues and pericocular encrustation. Statistically significant decreases in body weights and body weight gains were also seen in rats exposed to 4824 mg/m^3 of TEG. At 2011 mg/m^3 , there were statistically significant decreases in body weights in males from Exposure Day 5. Females from the 2011 mg/m^3 exposure concentration group and rats of both sexes from the 494 mg/m^3 exposure concentration group had body weights and body weight gains that were not significantly different from the controls. The most notable gross findings from TEG-exposed rats were unkempt fur, swollen eyelids with periocular and perinasal discharge and crusting, and multifocal or diffuse color change due to congestion and/or hemorrhage of various organs and tissues.

Rats from the 5000 mg/m^3 exposure concentration group all died or were sacrificed in a moribund condition on or before the beginning of Exposure Day 5. Therefore, only limited data were obtained from these animals. In addition to the gross pathology findings noted above, hyperinflation of the lungs (failure of lungs to collapse when the chest cavity was opened) was observed during necropsy in 5 males and 10 females from the 5000 mg/m^3 exposure concentration group. Ocular opacities were also seen in 5 males and 5 females from this exposure group. The most prominent microscopic lesions found in the 5000 mg/m^3 TEG-exposed rats which died or were sacrificed moribund involved congestion and, occasionally, hemorrhage of many organs and tissues. The pituitary, nasal mucosa, brain, and lungs were affected in many of the rats of both sexes. Congestion of the kidneys and hemorrhage in the thymus were also relatively common in the females.

For TEG-exposed rats that survived the exposure regimen, food consumption was statistically significantly increased in an exposure concentration-related fashion for females only at 494 and 2011 mg/m^3 . Water consumption was statistically significantly increased in both sexes at 2011 mg/m^3 and in females at 494 mg/m^3 . The only statistically significant hematological effects occurred in females from the 2011 mg/m^3 group which included slight increases in erythrocyte count and slight decreases in mean corpuscular volume. Notable clinical chemistry findings were increased activities of alanine aminotransferase at 2011 mg/m^3 and alkaline phosphatase at 494 and 2011 mg/m^3 , and slight increases in blood urea nitrogen and inorganic phosphorous in females from the 494 and 2011 mg/m^3 exposure concentration groups. Urinalysis showed statistically significant increases in urine volume and decreases in osmolality, pH, and N-acetyl- β -D-glucosaminidase activity at 2011 mg/m^3 , with a trend for changes in these values at 494 mg/m^3 . Absolute liver and kidney weights were increased in females from the 2011 mg/m^3 exposure concentration group and increased relative (as a percentage of body weight) weights were measured for both organs at 2011 mg/m^3 . There was no histological evidence of liver or kidney injury noted in animals from any exposure concentration group. The only microscopic lesion was minimal to mild

alveolar histiocytosis, which was in excess of that for the controls at 2011 mg/m³, but not at 494 mg/m³. The above findings possibly indicated a slight impairment of liver function, but without morphological evidence of organ injury.

This study demonstrated that daily inhalation exposures to TEG aerosol at or above concentrations of 5000 mg/m³ were fatally toxic to rats within 5 days. Similar exposures for up to 9 days at or below concentrations of 2000 mg/m³, however, did not result in life threatening signs of toxic effects. The urinary changes and associated increase in water consumption are consistent with an osmotic diuresis resulting from the excretion of absorbed TEG and its metabolites. There were no consistent findings suggestive of renal injury. The increased serum enzyme activities suggest minimal hepatotoxicity, which, for this study, has a threshold exposure concentration for this toxic response that is near to 494 mg/m³.

TARGET CONCENTRATION SELECTION

Target triethylene glycol aerosol concentrations of 0 (control), 100, 500, and 1000 mg/m³ were selected by the Sponsor based on the previous inhalation studies. In order to limit exposure to the inhalation route, and thus avoid unquantifiable dosage, it was considered appropriate to conduct the study by nose-only exposure.

MATERIALS AND METHODS

The protocol and any protocol amendments detailing the design and conduct of this study are included in Appendix 12. Protocol deviations are also included in Appendix 12.

Test Substance

Two 1-gallon plastic containers of triethylene glycol (TEG; Lot No. TS-2051109, CAS No. 112-27-6) were received on November 12, 1993, from Union Carbide Corporation, Texas City, TX and assigned BRRRC Sample No. 56-415-1 and 56-415-2. TEG was a clear liquid and was stored at room temperature. Identification of TEG was confirmed by gas chromatography/mass spectroscopy and nuclear magnetic resonance. Its purity was determined by the GLP Analytical Skill Center at the UCC South Charleston, WV, Technical Center to be 99.9%, and the report is included in Appendix 1.

Animals and Husbandry

Sixty-five male and 65 female CD⁰ rats arrived on November 15, 1993, from Charles River Laboratories, (Portage, MI). They were designated by the supplier to be approximately 35 days old (the birth date was recorded as October 11, 1993) upon arrival. The females were nulliparous and nonpregnant.

Animals were housed in Room 103 from arrival to termination of the study, except during exposures. Within 2 days of receipt, the animals were examined by a clinical veterinarian and a pretest health screen for representative animals was initiated. The health screen included full necropsy, histologic examination of selected tissues (including respiratory tract) and examinations for fecal parasites. Approximately 2 weeks later, serum viral antibody

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analyses were conducted on 5 animals/sex. Based on the results of these data, the clinical veterinarian indicated that these animals were in good health and suitable for use.

All animals were assigned unique numbers and identified by cage tags. Animals considered available for the study were also identified by a tail tattooing procedure. Animals selected for the pretest health screen were identified after sacrifice by a toe-clipping procedure.

The animals were individually housed in stainless steel, wire mesh cages (22.5 x 15.5 x 18.0 cm). DACB® (Deotized Animal Cage Board; Shepherd Specialty Papers, Inc.) was placed under each cage and changed regularly 2 or 3 times each week. Cages were changed and sanitized at least once every 2 weeks. An automatic timer was set to provide fluorescent lighting for a 12-hour photoperiod (approximately 0500 to 1700 hours for the light phase). Temperature and relative humidity were recorded (Cole-Parmer Hygrothermograph Seven-Day Continuous Recorder, Model No. 8362-00 Cole-Parmer Instrument Co., Chicago, IL). Temperature was routinely maintained at 66-77°F; relative humidity was routinely maintained at 40-70%. Any minor exceptions to these specified ranges were noted in the raw data.

Tap water (Municipal Authority of Westmoreland County, Greensburg, PA) was available ad libitum, except during exposures, and was delivered by 250 ml plastic water bottles. Water analyses were provided by the supplier, Halliburton NUS Environmental Laboratories, Professional Service Industries, Inc., Lancaster Laboratories, Inc. or Chester Lab and RJ Lee Group, Inc. at regular intervals. EPA standards for maximum levels of contaminants were not exceeded. Ground Lab Diet™ The Richmond Standard™ Certified Rodent Diet #5002 (PMI Feeds, Inc.) was available ad libitum, except during exposures. Analyses for chemical composition and possible contaminants of each feed lot were performed by PMI Feeds, Inc., and the results were included in the raw data.

Animal Acclimation

The acclimation period was approximately 3 weeks. During this period, the animals were weighed at least 2 times at scheduled intervals. Detailed clinical observations were conducted in conjunction with body weight measurements. Cage-side animal observations were conducted at least once daily, and mortality checks were conducted twice daily (morning and afternoon). The animals were examined just prior to the end of the acclimation period by a clinical veterinarian. Animals considered unacceptable for the study, based on the clinical signs, body weight, or body weight gain, were rejected. The fate of rejected animals, and the reasons for rejection were documented in the raw data.

Study Organization

Following the second pretest body weight, the animals were assigned to 3 exposure groups and a control group using a nonstratified randomization procedure based on body weight. At the time of group assignment, only animals with body weight within $\pm 20\%$ of the population mean for each sex were included. The body weight range on the day of first exposure was 257.8 to

332 g for males and 168.5 to 242 g for females. The following table summarizes the organization of the study.

Group	Number of Animals		Target Concentration (mg/m ³)
	Male	Female	
Control ^a	15	15	0
Low	10	10	100
Intermediate	10	10	500
High ^a	15	15	1000

^aFive animals/sex of the control and high concentration groups were assigned to the 4-week recovery group.

The exposures began on December 6, 1993 (Study Day 1). Animals were exposed for 6 hours/day for 5 consecutive days. The 6-hour exposure period was defined as the time when the animals were connected to and then disconnected from the exposure chamber. After 2 days without exposure (weekend), the animals were exposed for an additional 4 consecutive days. All control animals were exposed only to filtered air using the same exposure regimen. Ten animals/sex/group were sacrificed on December 17, 1993, the day after 9 exposures. The 5 animals/sex of the control and high concentration groups assigned to the recovery group were sacrificed on January 14, 1994, 4 weeks after the final exposure.

Exposure to the Test Substance

Inhalation Chamber Description and Operation

The inhalation chambers used for this study were located in Room 137. A polyvinyl chloride (PVC) exposure chamber was used. These exposure chambers utilize the flow-past, nonbreathing concept (Cannon *et al.*, 1983) for TEG exposure of the animals. The exposure chambers were composed of separate tiers, with each tier containing a total of 8 exposure ports for exposing 8 animals (1 exposure port/animal). Each animal was housed in a plexiglas tube (5.7 cm diameter tapered front x 19.5 cm length). The control and 1000 mg/m³ exposure chambers had 4 tiers, and the 100 and 500 mg/m³ exposure chambers had 3 tiers.

Chambers were provided with air at a flowrate of approximately 16 lpm for the 0 and 1000 mg/m³ and 12 lpm for the 100 and 500 mg/m³ to ensure an adequate oxygen content of at least 19%. The airflow rates were monitored continuously and recorded approximately every 30 minutes. All chambers were maintained at a slightly negative pressure.

Aerosol Generation

Liquid TEG was aerosolized, for all exposure levels, by positioning a single barrel Laskin Aerosol Generator (Enviro-Air Tech, Inc., Goshen, NY) into a glass 3-neck flask containing the TEG. The TEG aerosol was introduced to the

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nose-only exposure chamber by filtered compressed air passing through the nebulizer and flask and exhausting through 1-inch glass tubing connected to the inlet tube of the chamber. Operating air pressures for the nebulizers ranged from 4 to 6.25 psi.

Chamber Atmosphere Measurements

Chamber concentrations in each exposure chamber were determined 3 times during each exposure period by standard gravimetric techniques. A complete description of the analytical procedures is included in Appendix 1.

The particle size distribution was measured using a TSI Aerodynamic Particle Sizer (TSI Incorporated, St. Paul, MN) and was determined each day for all exposure groups. The data collected were analyzed by probit analysis (Hinds, 1982) to obtain the mass median aerodynamic diameter (MMAD) and the geometric standard deviation (σ_g).

Chamber temperatures were recorded using a K thermocouple connected to a digital recorder (Fluke 51 K/J Thermometer, John Fluke Mfg. Co., Inc., Everett, WA). Temperature measurements were recorded approximately 2 times each exposure hour.

Observations and Measurements

In-life Evaluations

All animals were individually observed for signs of toxic effects, except during the exposures. During the exposures, observations were recorded on a group basis. Preceding and following each exposure, observations were recorded for animals exhibiting overt clinical signs. At the time of body weight collection and just preceding sacrifice, detailed observations were performed on all animals. On nonexposure days, the animals were observed once a day for overt clinical signs and twice a day for mortality.

Body weights were measured for all animals on the morning prior to initiation of the first exposure (denoted as Study Day 1 in the tables), Study Days 2, 5, 8, 9, and immediately preceding sacrifice. The animals were weighed weekly during the 4-week recovery period. Body weight gains were calculated for the periods between weighings.

Food and water consumption were measured over Study Days 1-2, 2-5, 5-8, 8-10 (males) and 8-11 (females).

Prior to the first exposure, the eyes of all rats were examined by a veterinarian using direct ophthalmoscopy following dilation of eyes with MYDRIACYL® 1% (tropicamide 1.0%) Ophthalmic Solution. Following the eighth exposure for the female rats and following the ninth exposure for the male rats, the eyes were again examined by a veterinarian by direct ophthalmoscopy following dilation of eyes. An ophthalmology examination was not performed on animals held for the 4-week recovery period. Details of the ophthalmology examination procedures are included in Appendix 4.

Clinical Pathology Evaluations

Prior to the final sacrifice, blood was obtained from all surviving animals, with the exception of those designated for the recovery phase, for hematology and clinical chemistry determinations. Blood samples were collected by retroorbital bleeding from methoxyflurane anesthetized rats on Day 12. Rats were not fasted prior to bleeding. Feed was removed from all animal cages prior to the start of the blood collection period, but water was supplied ad libitum. Blood samples were not collected on the recovery animals.

Following the eighth exposure (Study Day 10) for male rats and following the ninth exposure (Study Day 11) for female rats, urine was collected over an approximate 18-hour period from all rats, except those designated for the recovery phase. The rats were housed individually in Nalgene® metabolic cages with stainless steel, wire mesh bottoms, approximately 22 cm diameter x 12 cm high (Nalge Company, Rochester, NY), during the collection period. Food and water were available ad libitum. Thymol crystals were added to the collection tube as a preservative. Urine samples were not collected on the recovery animals.

The following were measured or calculated:

Hematology

hematocrit	total leukocyte count
hemoglobin	differential leukocyte count
erythrocyte count	platelet count
mean corpuscular volume (MCV)	reticulocyte count
mean corpuscular hemoglobin (MCH)	
mean corpuscular hemoglobin concentration (MCHC)	

Clinical Chemistry

glucose (nonfasting)	aspartate aminotransferase (AST)
urea nitrogen	alanine aminotransferase (ALT)
creatinine	creatinine kinase (CK)
total protein	lactate dehydrogenase (LDH)
protein electrophoresis	gamma-glutamyl transferase (GGT)
albumin	sorbitol dehydrogenase (SDH)
globulins	alkaline phosphatase (ALK)
total bilirubin	
direct bilirubin	
indirect bilirubin (calculated)	
calcium	
phosphorus	
sodium	
potassium	
chloride	

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Urinalysis and Urine Chemistry

osmolality	blood
pH	urobilinogen
protein	total volume
glucose	color and appearance
ketones	microscopic elements
bilirubin	N-acetyl- β -D-glucosaminidase (NAG)
creatinine	alpha _{2u} -globulin
creatinine clearance (calculated)	

Details of the clinical pathology procedures are included in Appendix 3.

Anatomic Pathology Evaluations

At the end of the exposure regimen, all surviving animals, with the exception of those designated for the recovery phase, were anesthetized with halothane and were euthanized by severing the brachial vessels to permit exsanguination. On the day of sacrifice, body weight was obtained to allow expression of relative organ weights. A complete necropsy was performed on all sacrificed animals. The liver, kidneys, brain, adrenals, lungs, spleen and testes were weighed for all sacrificed animals. An additional 5 rats/sex/group from the control and high exposure groups were euthanized in the same manner following a 4-week recovery period. The recovery animals also received a complete necropsy. The following tissues were collected and retained in 10% neutral buffered formalin:

<u>gross lesions</u>	adrenals
<u>lung</u>	testes
<u>nasopharyngeal tissue</u>	ovaries
brain	<u>urinary bladder</u>
thymic region	lymph nodes
<u>trachea</u>	mesenteric
heart	non-mesenteric
<u>liver</u>	nerve, sciatic
spleen	eyes
<u>kidneys</u>	<u>larynx</u>

Tails were saved for identification purposes.

The underlined tissues from all animals of the control and high exposure groups from the Day 12 sacrifice were processed histologically and examined microscopically. In addition, the lungs, liver, kidneys, and all gross lesions for all animals from the low and intermediate exposure groups were examined.

Details of the anatomic pathology procedures are included in Appendix 2.

Data Analyses

The data for quantitative continuous variables were intercompared for the 3 exposure groups and the control group by use of Levene's test for equality of

variances, analysis of variance (ANOVA), and t-tests. The t-tests were used when the F value from the ANOVA was significant or only one group was compared to the control group. When Levene's test indicated similar variances, and the ANOVA was significant, a pooled t-test was used for pairwise comparisons. When Levene's test indicated heterogeneous variances, all groups were compared by an ANOVA for unequal variances followed, when necessary, by a separate variance t-test for pairwise comparisons.

Nonparametric data were statistically evaluated using the Kruskal-Wallis test followed by the Mann-Whitney test. Incidence data were compared using Fisher's Exact Test. For all statistical tests, the probability value of < 0.05 (two-tailed) was used as the critical level of significance.

Various models of calculators, computers, and computer programs may have been used to analyze data for this study. Since various models round or truncate numbers differently, values in some tables may differ slightly from those in other tables or from independently calculated data. The integrity of the study and interpretation of the data were unaffected by these differences.

RETENTION OF RECORDS

All raw data, documentation, the protocol and any amendments, specimens, and a copy of the final report generated as a result of this study will be retained in the BRRRC Archives for at least 5 years. A reserve sample of TEG from each container used during the study will also be stored in the BRRRC Archives.

RESULTS AND DISCUSSION

All references of differences in group mean values in the following text refer to comparisons of statistically significant differences between the exposure group and the control group, unless otherwise noted. Repeated reference to the control group and the statistical significance will not be made in order to simplify the text.

Chamber Atmosphere

Detailed results and discussion of the chamber atmosphere measurements are included in Appendix 1.

Mean chamber gravimetric concentrations (\pm SD) were 102 (\pm 8.2), 517 (\pm 37.9), and 1036 (\pm 27.2) mg/m^3 for the target concentrations of 100, 500, and 1000 mg/m^3 , respectively. The mean analytical/nominal (A/N) concentration ratios were 0.84, 1.19, and 1.08 for TEG target concentrations of 100, 500, and 1000 mg/m^3 , respectively. These A/N values are typically low for aerosol studies, possibly due to losses of the aerosol on chamber walls, animal fur, animal cages, and generation equipment. The nominal concentration was not calculated for Exposure Day 9, since only the start time for the exposure was recorded. The mean nominal concentrations (\pm SD) were 132 (\pm 37.1), 454 (\pm 95.0), and 968 (\pm 87.8) mg/m^3 for the target concentrations of 100, 500, and 1000 mg/m^3 , respectively. The mean MMADs were 1.2, 1.4, and 1.3 microns for the 100, 500, and 1000 mg/m^3 groups, respectively, with the daily Gg ranging from 1.25 to 1.39 for all groups. The mean chamber temperature range for all exposures was 19.5 to 20.8°C.

Clinical Observations and Mortality

Summaries of the clinical observations are presented in Tables 1 and 2. Individual animal clinical observation data are included in Appendix 5.

One male in the control group died during the first exposure and was replaced with another animal. One male and 1 female were found dead in the 500 mg/m³ group on Days 1 and 2, respectively. One male in the 0 mg/m³ group was observed with alopecia (Day 18), periocular encrustation around either the right eye or both eyes (Days 8-40), and swollen periocular tissue around either the right eye or both eyes (Days 8-40). One male in the 500 mg/m³ group was observed to have trauma to the right side of its body (Day 9). Unkempt fur (Day 5) was observed in 1 male from the 1000 mg/m³ group. Another male from the 1000 mg/m³ group was observed with ocular discharge of the left eye (Day 25), swollen periocular tissue of the left eye (Day 25), malocclusion (Days 18-25), and a broken incisor (Day 18). These deaths and clinical findings were not considered related to the TEG exposure. No clinical signs were observed in the female rats.

Body Weights

Summaries of absolute body weight and body weight gain are presented in Tables 3 to 6. Individual animal body weight data are included in Appendix 6.

No changes in body weights for the males or females or body weight gains for the males in any TEG exposure groups were noted. A nonstatistically significant decrease in female body weight gains in the 500 and 1000 mg/m³ groups was observed on Days 8 to 9 and 9 to 12.

Food Consumption

Summaries of food consumption data are presented in Tables 7 and 8. Individual animal food consumption data are included in Appendix 7.

No changes in food consumption for the males or females in any TEG exposure groups were noted.

Water Consumption

Summaries of water consumption data are presented in Tables 9 and 10. Individual animal water consumption data are included in Appendix 8.

No changes in water consumption for the males or females in any TEG exposure groups were noted.

Ophthalmology Examinations

Individual animal ophthalmology data are included in Appendix 11. Detailed results and discussion of the ophthalmology examinations are included in Appendix 4.

No ophthalmic lesions found in this study were attributed to TEG exposure.

Clinical Pathology Evaluations

Summaries of the hematology measurements are presented in Tables 11 and 12. Summaries of the clinical chemistry measurements are presented in Tables 13 and 14. Summaries of the protein electrophoresis measurements are presented in Tables 15 and 16. Summaries of the urine chemistry measurements are presented in Tables 17 and 18. Summaries of the urinalysis measurements are presented in Tables 19 and 20. Individual clinical pathology data are included in Appendix 10. Detailed results and discussion of the clinical pathology measurements are included in Appendix 3.

There were no exposure-related differences for hematology, serum chemistry, protein fractions, urine chemistry, and urinalysis determinations for male and female rats in any TEG exposure group.

Organ Weights, Necropsy Observations, and Microscopic Diagnoses

Summary results of organ weights, organ weights relative to final body weight, and organ weights relative to brain weight are presented in Tables 21 to 32. Summary results of necropsy observations are presented in Tables 33 to 38. Summary results of microscopic diagnoses are presented in Tables 39 to 42. Individual anatomic pathology data are included in Appendix 9. Detailed results and discussion of the anatomic pathology results are included in Appendix 2.

At the Day 12 sacrifice, no organ weight changes in the males or females were observed. At the Day 40 sacrifice, the absolute and relative (as percentages of body and brain weights) adrenal gland weights were increased in the males in the 1000 mg/m³ group. The relative (as a percentage of body weight) adrenal gland weights in the females increased in the 1000 mg/m³ group. As the organ weight changes occurred 4 weeks after the TEG exposures were terminated, they were not considered related to TEG.

No gross or microscopic lesions noted in the TEG-exposed animals were believed related to TEG exposure.

CONCLUSIONS

The purpose of this study was to determine and evaluate the toxic effects in rats resulting from a 6-hour repeated nose-only exposure to a TEG aerosol for 9 days.

The only finding apparently related to the TEG exposures was the nonstatistically significant decrease in female body weight gains in the 500 and 1000 mg/m³ groups over Days 8 to 9 and 9 to 12.

Based on the small decreased body weight gains for the females in the 500 and 1000 mg/m³ group, a no-observed-effect level (NOEL) was determined to be 100 mg/m³ for a 9-day repeated nose-only exposure to a TEG aerosol in rats.

REVIEW AND APPROVAL

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Senior Manager:

Heather D. Burleigh-Flayer 10/26/94
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Additional personnel are listed in the raw data.

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