TSCA HEALTH & SAFETY STUDY COVER SHEET 8EHQ-0699-14328

TSCA CBI STATUS:

-CHECK IF THIS PAGE CONTAINS CONFIDENTIAL BUSINESS INFORMATION (CBI)

Clearly mark the confidential information with bracketing and check the box in the appropriate section (☐ *Contains CBI*).
Submit a sanitized cover sheet with CBI deleted. Mark the sanitized copy, "Public Display Copy" in the heading.

1.0 SUBMISSION TYPE --*Contains CBI*

☐ 8(d) **X 8(e)** ☐ FYI ☐ 4 ☐ OTHER: Specify_____

- Intial Submission XX -Follow-up Submission -Final Report Submission

Previous EPA Submission Number or Title if update or follow-up: Docket Number, if any: #

8(e)HQ-98-14328 RDCN 889900000 47

☐ continuation sheet attached

2.1 SUMMARY/ABSTRACT ATTACHED (may be required for 8(e): optional for §4, 8(d) & FYI) X - YES ☐ NO	**2.2 SUBMITTER TRACKING NUMBER OR INTERNAL ID** Cert# P 917006913 99-2-32	**2.3 FOR EPA USE ONLY**

3.0 CHEMICAL/TEST SUBSTANCE IDENTITY -*Contains CBI*

CAS#: 111988-49-9

Reported Chemical Name (specify nomenclature if other than CAS name): (Cyanamide, [3-(6-chloro-3-pyridinyl)methyl]-2]thiazolidinylidene]-,

Purity _____%

X – Single Ingredient

☐ Commerical/Tech Grade

☐-Mixture *Trade Name:* YRC 2894 *Common Name:* Chlornicotinyl

1999 JUN 18 AM 10:50 RECEIVED OPPT CBIC

4.0 REPORT/STUDY TITLE - *Contains CBI*

Study on acute Inhalation Toxicity in Rats According to OECD # 403, Report # 28115-A

☐ Continuation sheet attached

5.1 STUDY/TSCATS INDEXING TERMS

[CHECK ONE]

HEALTH EFFECTS (HE): X ENVIRONMENTAL EFFECTS (EE):_____ ENVIRONMENTAL FATE (EF):_____

5.2 STUDY/TSCATS INDEXING TERMS (see instructions for 4 digit codes)

STUDY TYPE: ATOX	SUBJECT ORGANISM (HE, EE only): RATS	ROUTE OF EXPOSURE (HE only):____	VEHICLE OF EXPOSURE (HEonly)_
Other:	Other:	*Other:*	*Other:*

6.0 REPORT/STUDY INFORMATION ☐ *Contains CBI* X- Study is GLP

Laboratory Bayer Toxicology, Wuppertal, Germany Report/Study Date: 4/24/99

Source of Data/Study Sponsor (if different than submitter) _____ Number of pages : 102

☐ continuation sheet attached

7.0 SUBMITTER INFORMATION ☐ *Contains CBI*

Submitter: Donald W. Lamb, Ph.D Title: V. P., Prod. Safety & Reg. Affrs Phone: 412-777-7431

Company Name: Bayer Corporation Company Address: 100 Bayer Road

Pittsburgh, PA 15205-9741 Submitter Address (if different): _____

Technical Contact: Donald W. Lamb, Ph.D Phone: (412)777-7431

☐ continuation sheet attached

99 JUL -9 AM 10:10 RECEIVED OPPT NCIC

8.0 ADDITIONAL/OPTIONAL STUDY COMMENTS ☐ *Contains CBI*

This compound is an experimental pesticide.

Contains No CBI

☐ continuation sheet attached

Submitter Signature Donald W. Lamb **Date:** 6/14/99

EHQ-98-14328 89990000233

9.0 CONTINUATION SHEET

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Submitter Tracking Number/Internal ID

P917006913
99-2-32

CONTINUED FROM COVER SHEET SECTION # 2.1

Abstract

The aerosol concentration in this study has been recalculated based on gravimetric analysis, but considering only non-volatile constituents of YRC 2894. Based on this change in the measurement of the aerosol concentration, the following changes have been made to the study summary:

Original Study Summary:

A study to evaluate the acute inhalation toxicity of YRC 2894 480 SC 05776/0096 in male and female Wistar rats was conducted. Groups of rats were exposed nose-only to average liquid aerosol concentrations of 690 mg/m3 and 1535 mg/m3 air. The results are summarized as follows:

Rat LC50 : male >1535 mg/m3 female >690<1535 mg/m3

male & female NOAEL: <690 mg/m3 air

A concentration of 690 mg/m3 was tolerated without mortality in both sexes

Revised Study Summary (changes from original text are bolded):

A study to evaluate the acute inhalation toxicity of YRC 2894 480 SC 05776/0096 in male and female Wistar rats was conducted. Groups of rats were exposed nose-only to average liquid aerosol concentrations of **989** mg/m3 and **2199** mg/m3 air. The results are summarized as follows:

Rat LC50 : male >**2199** mg/m3 female >**989**<**2199** mg/m3

male & female NOAEL: <**989** mg/m3 air

A concentration of **989** mg/m3 was tolerated without mortality in both sexes.

SUPPLEMENTAL SUBMISSION TO AC NO. 108683

STUDY TITLE

YRC 2894 480 SC 05776/0096
(c.n.: Thiacloprid)
Study on Acute Inhalation Toxicity in Rats
According to OECD No. 403

DATA REQUIREMENT

US EPA-FIFRA Guideline No. 81-3

AUTHORS

108683-1

Priv.-Doz. Dr. J. Pauluhn
and
Dr. M. Temerowski

FILE
8689

STUDY COMPLETION DATE

Original Report: November 4, 1998
Supplemental Report: April 24, 1999

PERFORMING LABORATORY

BAYER AG
DEPARTMENT OF TOXICOLOGY
Friedrich-Ebert-Strasse 217-233
D-42096 Wuppertal
Germany

LABORATORY PROJECT ID

Bayer AG Report No. 28115 A
Bayer AG Study No. T 6067418

STATEMENT OF DATA CONFIDENTIALITY

No claim of confidentiality is made for any information contained in this study on the basis of its falling within the scope of FIFRA Section 10(d)(1)(A), (B), or (C):

BAYER CORPORATION

Dr. J.H. Thyssen: _____
Vice President, Toxicology

Date: _____

GOOD LABORATORY PRACTICE STATEMENT

This study was conducted in compliance with the OECD Principles of Good Laboratory Practice (GLP) and with the Principles of Good Laboratory Practice (GLP) according to Annex 1 ChemG (1994; *cf.* references) and meets the FIFRA Good Laboratory Practice Standards (40 CFR Part 160), with the exception that recognized differences exist between the GLP principles/standards of OECD and FIFRA (for instance, authority granted Agency inspectors and certain record retention requirements).

Date: April 24, 1999

Dr. J. Pauluhn D.A.B.T.
Board Approved Toxicologist (DGPT)
Study Director

SPONSOR:

BAYER AG

Date: May 02, 1999

Dr. B. Stahl
Project Management

SUBMITTER:
BAYER CORPORATION

Date: _____

Dr. J.H. Thyssen
Vice President, Toxicology

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QUALITY ASSURANCE STATEMENT

Test Item: **YRC 2894 480 SC 05776/0096**

Study No.: **T6067418**

Study-based inspections/audits were conducted by the Quality Assurance on the dates given below. Audit reports have been submitted in writing to the study director and, if necessary, also the laboratory management, or other persons affected.

Date of audit		Date of report to study director and/or management
Sep. 10, 1998	(study plan)	Sep. 10, 1998
Sep. 14, 1998	(study conduct)	Sep. 14, 1998
Oct. 22, 1998 - Oct. 23, 1998	(first draft)	Oct. 23, 1998
Mar. 25, 1999	(final draft)	Apr. 09, 1999

The results of this revised report and the methods used have been correctly reported.

Quality Assurance Unit
PH-OP-QA-GLP, Bayer AG

Date: Apr. 16, 1999

Responsible: Dr.R.Rauchschwalbe

3 Signatures

Study director: _____ Date: 29.4.99
(PD Dr. J. Pauluhn)

Report author: _____ Date: 15.04.1999
(Dr. M. Temerowski)

Institute Head: _____ Date: April 26, 99
(Prof. Dr. G. Schlüter)

4 Summary and Assessment

A study on the acute inhalation toxicity of YRC 2894 480 SC 05776/0096 (hereafter referred to as *test substance*) on rats has been conducted in accordance with OECD Guideline No. 403. Groups of rats (male and females) were nose-only exposed to an average liquid aerosol concentration of 989 mg/m³ and 2199 mg/m³ air. Attempts were made so that aerosol generated was respirable to rats. The results can be summarized as follows:

LC50 inhalation (aerosol, 4 hr)	NO(A)EL
Males > 2199 mg/m³ [1)]	Males & females: < 989 mg/m³ air[1)].
Females: > 989 mg/m³ - < 2199 mg/m³ [1)]	

Observations and Measurements: Exposure to the maximum tested concentration of 2199 mg/m³ was accompanied with mortality in female animals only and - in both sexes - with clinical signs indicative of respiratory tract irritation (bradypnea, labored breathing pattern, dyspnoea, nostrils reddened and encrusted), hypothermia, piloerection, salivation, pallor, stilted gait, increased muscle tone, retardation in body weight gain and central nervous effects (tremor, reduced motility, atony, dilated pupils). The duration of signs appeared to be governed by respiratory signs such as laboured breathing (until day 13) and unspecific signs such as piloerection (until day 7). A concentration of 989 mg/m³ was tolerated without mortality in both sexes but with few symptoms, mainly due to respiratory tract irritation (bradypnea, labored breathing pattern), retardation in body weight gain and central nervous effects (reduced motility and tremor). No treatment-related findings were detected in the rats sacrificed at the end of the 2-week postexposure period or found dead.

[1] This concentration represents the actual concentration of the test substance in the rats' breathing zone.

With regard to the respirability of the aerosol generated internationally recognized recommendations such as of SOT (1992) were almost fulfilled, i.e. the MMAD was less than 4 µm in the low-dose groups (MMAD ≈ 2,88 µm, GSD ≈ 1.66) and slightly above 4 µm in the high-dose group (MMAD ≈ 4,18 µm, GSD ≈ 2,06).

In summary, the aerosolized test substance (liquid aerosol) proved to have a moderate acute inhalation toxicity to female rats and a low acute inhalation toxicity to male rats. Specific pathognomonic findings indicate a causal relationship between exposure concentration and signs related to respiratory tract irritation.

5 Introduction

This acute inhalation toxicity study was conducted in accordance with OECD Guideline No. 403 and the respective EU-Guideline using YRC 2894 480 SC 05776/0096 as a test substance. The study was performed on rats (nose-only exposure over 4 hours to a liquid aerosol, dynamic exposure conditions, 2 week observation period). This study served the purpose of product classification and to estimate the potential acute health hazard resulting from handling this product.

Testing facility:

Institute of Toxicology - Industrial Chemicals/Department of Occupational Toxicology, Bayer AG, D-42096 Wuppertal, Friedrich-Ebert-Straße 217 - 333, Germany.

Study/project identification:

Study no.: T 6067418

Study period:

September 14, 1998 - September 30, 1998
Experimental starting date: September 11, 1998 (technical pre-trials)
Study completion date: see signature of study director (page 7)

Sponsor:

BAYER AG, Agriculture Division

6 Responsibilities

Air conditioning/air make-up:	Dipl. Ing. G. Strietholt
Archiving of study data:	Prof. Dr. G. Schlüter
Biometric evaluation:	Dr. J. Pauluhn
Gross pathology:	Dr. Rosenbruch
Head of Department:	Prof. Dr. G. Schlüter
Laboratory Animal Services:	Dr. Petersen v. Gehr
Quality Assurance:	Dr. H. Lehn
Study Director:	Dr. J. Pauluhn
Report Author:	Dr. M. Temerowski
Analytical Chemistry:	Dr. M.G. Teller

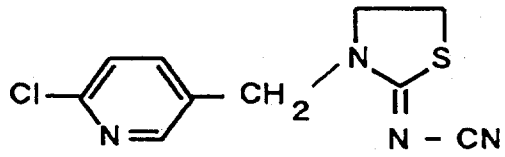
7 Materials and Methods

7.1 Test Substance

Test substance:	YRC 2894 480 SC 05776/0096
Formulation no. (batch no.):	0100 based on formulation no. 05776/0096
Developmental No.:	0215336
Indication:	insecticide
Content:	41.3 % YRC 2894
Specific density:	1,188 g/cm^3 at 20 °C
Manufacturer:	Bayer AG
Approval:	until November 05, 1998
Appearance:	white to pale-beige
Storage:	room temperature

Active ingredient

Common name:	Thiacloprid
Chemical name (C.A):	Cyanamide, [3-[(6-chloro-3-pyridinyl)methyl]-2-thiazolidinylidene]-
CAS Registry no.:	111988-49-9
Molecular weight:	252.8 g/mol
Molecular formula:	$C_{10}H_9ClN_4S$
Structural formula:	



7.2 Test System and Housing of Animals

Species and species justification: The study was carried out in rats, a rodent species recommended in the test guidelines.

Healthy young adult SPF bred Wistar rats, strain Hsd Cpb:WU (SPF), from the experimental animal breeder Harlan, Winkelmann, Borchen (Germany), were used. Animals of this strain have been used at Bayer AG in toxicological studies for years. Historical data on their physiology, diseases and spontaneous alterations are available. The state of health of the strain is randomly checked regularly at the instance of the Laboratory Animal Services, Bayer AG, for the most important specific infectious pathogens. The results of these examinations are archived.

Acclimatization: The animals were acclimatized to the animal room conditions for at least 5 days before use.

Identification: Animals were identified by both individual color-marking and cage-labels. All animals from this study were located on one cage-rack.

Randomization: Before the start of the study the health status of each animal was assessed. Animals were subsequently assigned to exposure groups at random (randomization procedure is described in section 7.17).

Health status: Only healthy rats free of signs were used for this study. The animals were not vaccinated or treated with anti-infective agents either before their arrival or during the acclimatization or study periods. The females were nulliparous and not pregnant.

Age and weight: At the study start the variation of individual weights did not exceed ± 10 per cent of the mean for each sex (see Appendix). Animals of the weight class used are about 7 (males) to 8-9 (females) weeks old and hence fulfill the criterion for young adults.

Animal housing: During the acclimatization and study periods the animals were housed singly in conventional Makrolon® Type II cages (based on A. Spiegel and R. Gönnert, Zschr. Versuchstierkunde, 1, 38 (1961) and G. Meister, Zschr. Versuchstierkunde, 7, 144-153 (1965)). Cages and water bottles were changed twice

a week while unconsumed feed was changed once per week. The legal requirements for housing experimental animals (86/609 EEC) were followed.

Bedding: Bedding consisted of type S 8/15 low-dust wood granulate from Ssniff, Soest/Westfalen, Germany. The wood granulate was randomly checked for harmful constituents at the request of the Laboratory Animal Services, Bayer AG.

Animal rooms: All animals were housed in a single room. For reasons of space availability rats from other acute toxicity studies were housed in the same room, however mistakes in animal assignments were excluded by adequate spatial separation, clear cage labeling, and appropriate organization of all work procedures. The housing of several studies in one animal room is not considered to be a deviation from current GLP-requirements since many acute studies comprise of 10 animals only (as required to perform a limit test).

Environmental Conditions in the Animal Room

The animal room environment was as follows:

Room temperature:	22 ± 2 °C
Relative humidity:	approximately 50 %
Dark/light cycle:	12 h/12 h; artificial light from 6.00 a.m. to 6.00 p.m. Central European Time
Light intensity:	approximately 14 watt/m² floor area
Ventilation:	approximately 10 air changes per hour

The room humidity and temperature were continuously monitored and documented using a calibrated thermohygrograph. Occasional deviations from these conditions occurred, e.g. as a result of animal room cleaning, but these had no detectable influence on the outcome of this study.

Cleaning, disinfection, and pest control: The animal room was regularly cleaned and disinfected once a week with an aqueous solution of TEGO® 2000. Contamination of the feed and contact with the test system were excluded. Pest control was not conducted in the animal room.

Feeding: Ration consisted of a standard fixed-formula diet (Altromin® 1324 pellets maintenance diet for rats and mice, Altromin GmbH, Lage) and tap water (drinking bottles). Both food and water were available *ad libitum*. The pelletized feed was contained in a rack in the stainless-steel wire cage cover. The nutritive composition and contaminant content of the standard diet was checked regularly by random sampling by the Laboratory Animal Services, Bayer AG. Details concerning general feed and water specifications are provided in the Appendix.

Water: Drinking quality tap-water (Drinking Water Decree of 05.12.1990, Bundesgesetzblatt [federal law gazette] part I, page 2612) was provided *ad libitum* in polycarbonate bottles containing approximately 300 ml (based on A. Spiegel and R. Gönnert, Zschr. Versuchstierkunde, 1, 38 (1961) and G. Meister, Zschr. Versuchstierkunde, 7, 144-153 (1965)). The results of feed and water analyses are retained by Bayer AG. The available data provided no evidence of an impact on the study objective.

7.3 Test Guidelines

The study described below was carried out in accordance with OECD Guideline No. 403. The study conditions were adjusted so as to fulfill both the EC Guideline 92/69/EEC and the FIFRA § 81-3 (US EPA, 1984) guideline. Other published recommendations (US EPA, 1988; SOT, 1992; Pauluhn *et al.*, 1996) were also taken into account so as to comply with internationally recognized procedures.

7.4 Exposure Conditions

Mode of exposure: Animals were exposed to the aerosolized test substance in Plexiglas exposure tubes applying a *directed-flow* nose-only exposure principle. Tubes were chosen that accommodated the animals size. These tubes were designed so that the rat's tail remained outside the tube, thus restrained-induced hyperthermia can be avoided. This type of exposure is preferable to whole-body exposure on scientific (Pauluhn, 1984) and technical reasons (rapid attainment of steady-state concentrations, no problems with regard to test atmosphere inhomogeneities, better capabilities to control all inhalation chamber parameters, easier cleaning of exhaust air, and lower consumption of test substance). Moreover,

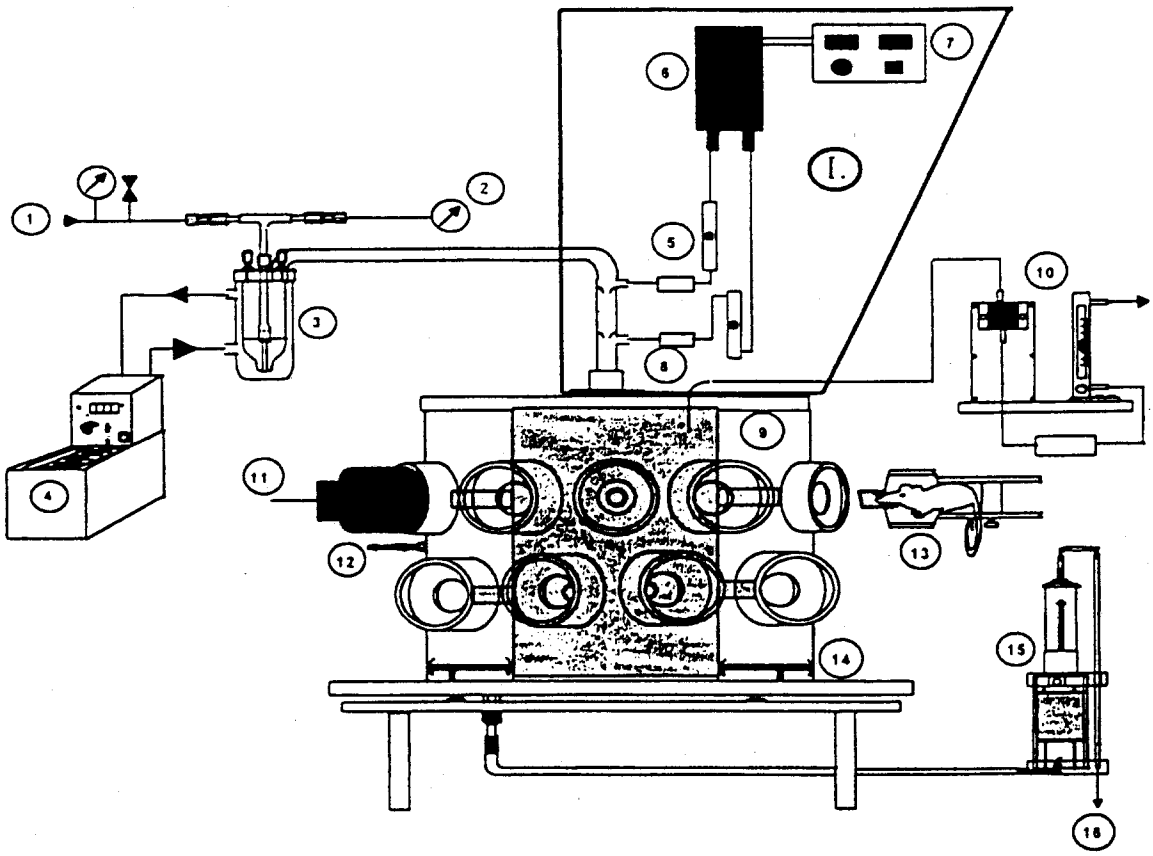
contamination of the fur can largely be avoided. The chambers used are commercially available (TSE, 61348 Bad Homburg) and the performance of this type of chamber has been published (Pauluhn, 1984; Pauluhn, 1988; Pauluhn, 1994).

Vehicle: The test substance was aerosolized as a 50% aqueous solution containing Levatit water as vehicle.

7.5 Aerosol Generation and Exposure Technique

Aerosol generation/aqueous solution: The liquid was nebulized using a *Collison nebulizer* with conditioned compressed air (15 litres of air/min; dispersion pressure approximately ≈150 kPa; for details see Table 1, result section). The temperature of the solution was adjusted to 20 °C via a Julabo waterbath prior to nebulization. For generation of low-dose test atmospheres, the test substance concentration was reduced via by-passing of 50% of the primary air flow, purified by a cotton-wool-filter and re-delivered to the primary air stream (for details see figure 1).

Figure 1: Inhalation Chamber (schematic)



1. Compressed conditioned air supply 2. Manometer 3. Collison-Nebulizer 4. Julabo-Thermostate **I. No. 5.-8. for 750 mg/m³ only** 5. Extraction of air with cotton-wool filter and Rotameter 6. Pump 7. Digital control unit 8. Dilution air supply with cotton-wool filter and Rotameter	9. Exposure zone 10. Real-time aerosol photometer 11. Sensor for temperature/humidity measurements 12. Sampling location ('breathing zone sampling') 13. Directed-flow nose-only exposure of rats in restrainer 14. Extraction of atmosphere from the chamber 15. Cotton-wool aerosol filter + HEPA filter 16. Exhaust air

Inhalation Chamber: The aluminum inhalation chamber has the following dimensions: inner diameter = 14 cm, outer diameter = 35 cm (two-chamber system), height = 25 cm (internal volume = about 3.8 l). The construction of the inhalation chamber is shown schematically in Figure 1. Details of this modular chamber and its validation with regard to spatial homogeneity of material distribution have been published (Pauluhn, 1994).

Conditioning the compressed air: Compressed air was supplied by Boge compressors and was conditioned (i.e. freed from water, dust, and oil) automatically by a VIA compressed air dryer. Adequate control devices were employed to control supply pressure.

Inhalation chamber steady-state concentration: The test atmosphere generation conditions provide an adequate number of air exchanges per hour (> 200 x, continuous generation of test atmosphere). Under such test conditions steady state is attained within the first minute of exposure ($t_{99\%}$ = 4.6 x chamber volume/flow rate; McFarland, 1976). The ratio between the air supplied and exhausted was chosen so that approximately 80-90% of the supplied air is removed via the exhaust system. The remainder provides adequate dead-space ventilation for the exposure tubes. At each exposure port a minimal air flow rate of 0.75 l/min was provided. The test atmosphere can by no means be diluted by bias-air-flows. The inhalation chamber was operated in a well ventilated chemical fume hood.

Air flows: During the exposure period air flows were monitored continuously and, if necessary, readjusted to the conditions required. Air flows were measured with calibrated flowmeters and/or soap bubble meter (Gilibrator, Ströhlein Instruments, Kaarst) and were checked for correct performance at regular intervals.

Treatment of exhaust air: The exhaust air was purified via cotton-wool and HEPA filters. These filters were disposed of by Bayer AG.

7.6 Inhalation Chamber Temperature and Humidity

The temperature and humidity measurements were made using a computerized system (Leybold Heraeus). These temperature and humidity sensors were located in the breathing zone area of the rats and in the exhaust location, respectively. The values were recorded at intervals of 10 min.The humidity sensors were calibrated using saturated salt solutions according to Greenspan (1977). The temperature sensors were calibrated with a calibration thermometer. Details of this type of monitoring have been reported elsewhere (Pauluhn, 1986).

7.7 Analysis of the Test Atmosphere

Nominal concentration: The nominal concentration was calculated from the ratio of the weight reduction of the collision nebulizer after cessation of the aerosol generation and the total throughput of air through the inhalation chamber. Specific information concerning air-flows and test atmosphere concentrations are provided in Table 1.

Gravimetric evaluation: The test-substance concentration was determined by gravimetric analysis (filter: Glass-Fibre-Filter, Sartorius, Göttingen, Germany; digital balance).

Samples were taken to allow direct comparison with cascade impactor analyses. All analytical concentrations reported refer to mg of test substance/m^3 air.

7.8 Characterization of Aerodynamic Particle-Size Distribution

The samples for the analysis of the particle-size distribution were also taken in the vicinity of the breathing zone. During the exposure two samples were taken.

The particle-size distribution was analyzed using a BERNER-TYPE AERAS low-pressure critical orifice cascade impactor (Hauke, Gmunden, Austria). Specifications and evaluations are provided in the Appendix. The individual impactor stages had been covered by an aluminum foil covered with a glass fibre filter which were subjected to gravimetric analysis. An adhesive stage coating (silicone spray) was not used to prevent particle bounce and re-entrainment, respectively, due to the physicochemical properties of the test substance. Gravimetrical analyses were made using a digital Sartorius M3P balance.

Evaluation of particle-size distributions: For the evaluation of the cascade impactor analyses the mass median aerodynamic diameter (MMAD) and the geometric standard deviation (GSD) are determined from the probit-transformed

cumulative particle mass frequency distribution (y-axis) and the logarithmic effective cut-off diameters (ECD's) (x-axis) of the individual impactor stages by linear regression. The GSD is calculated from the regression line: percentile 84 / percentile 50. The relative mass with an aerodynamic diameter ≤ 3 µm ("respirable mass fraction") [Raabe, 1982; Snipes, 1989; SOT-Commentary, 1992] is calculated from the regression line. For probit transformation and linear regression FORTRAN algorithms published by Rosiello *et al.* (1977) are used. The MMAD was calculated using published following formulas [Marple and Rubow, 1980; Pauluhn, 1994].

To verify whether the aerosol distribution is in fact unimodal and log-normal the normalized mass per stage (f_H') is evaluated as a histogram. $\Delta \log D_p$ is equal the difference $\log D_{p+1}$ - $\log D_p$, whereas D_p is the lower (left) cut-size limit and D_{p+1} the higher (right) cut-size limit of the corresponding impactor stage. As demonstrated by the evaluations included in the Appendix, the impactor stage cut-off limit (D_{p+1}) to the right was used for all calculations.

$$f_{H}' = \frac{1}{N_f} \times \frac{mass\ /\ stage}{\Delta \log D_p}$$

The log-normal mass distribution $y'(D_{ae}) = 1/N_f \times y(D_{ae})$ as a function of the aerodynamic diameter (D_{ae}) is computed using the formula:

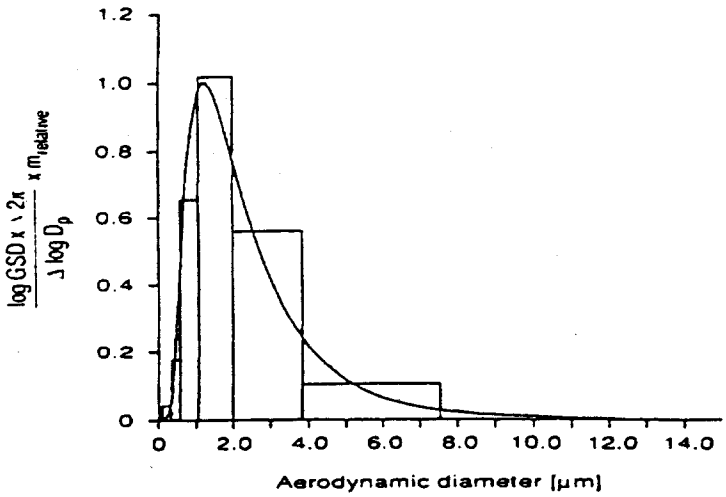
$$y'(D_{ae}) = \exp\left[-\frac{(\log D_{ae} - \log MMAD)^2}{2 \times \log^2 GSD}\right]$$

The normalization factor (N_f) is calculated as follows:

$$N_f = \frac{\Sigma mass}{\log GSD \times \sqrt{2\pi}}$$

Where Σmass is the total mass collected by the cascade impactor, where $m_{relative}$ = mass per stage/Σmass (*cf.* Fig. 2).

Figure 2: Principle of characterization of aerosol atmosphere

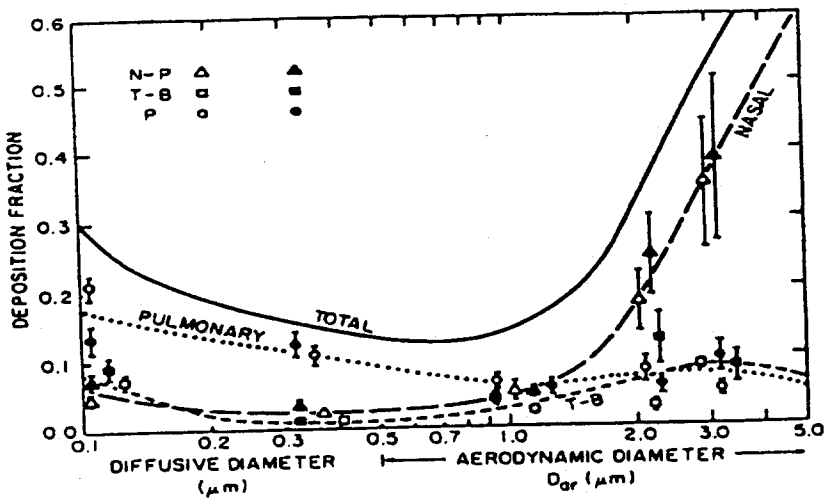


The algorithm for the calculation of particle size characteristics is taken from pertinent reference works on aerosol physics (Dennis, 1976; Marple and Rubow, 1980) and proves to be generally applicable (Pauluhn 1988; Pauluhn, 1994).

Respirability

Fig. 3 below, demonstrates that the particle-size distribution achieved is adequate to reach all potential target structures of the respiratory tract.

Figure 3: Respirability of Aerosols (Raabe, 1982)



7.9 Collection Efficiency

The sampling equipment was adjusted with calibrated rotameters to internationally recognized standards (ACGIH, 1978; Section I "Calibration of Air Sampling Instruments").

The conditions for generating the test atmosphere are optimized to provide maximum aerosol respirability to rats (Raabe, 1982; Snipes, 1989; SOT-Commentary, 1992). The absence of larger particles and high flow rates in the vicinity of the sampling ports make it possible to disregard potential anisokinetic sampling errors, thus ensuring a representative sampling even with different sampling probe orifice diameters and flow rates. The tolerance limits for the radius of the probe orifice are calculated using the following formula [ACGIH, 1978].

Calculations consider both a particle size distribution that encompasses aerodynamic diameters (D_{ae}) of 0.5 to 7.4 µm and sample flows ranging from 8 to 80 ml/sec.

$$5 \times \sqrt[3]{\frac{flow \times \tau}{4 \times \pi}} \leq r_p \leq \frac{1}{5} \times \sqrt[2]{\frac{flow}{g \times \tau \times \pi}}$$

r_p = radius of the sample probe in cm = ½ x D_p

τ = relaxation time ($D_{ae\ 0.5\ \mu m}$ = 1×10^{-6} sec; $D_{ae\ 7.4\ \mu m}$ = 1.7×10^{-4} sec)

g = gravity constant = 980 cm/sec^2

Tolerance limits calculations for the sample probe orifice (r_p) indicated that a representative sampling is assured when the orifice inner diameter is in the range of 1.0 to 1.6 cm. Orifices of the sampling instruments used here are in compliance with this criteria. Details of the D_p tolerance limit calculations are published elsewhere (Pauluhn, 1988; Pauluhn, 1994).

7.10 Stability of the Test Atmosphere

The integrity and stability of the aerosol generation and exposure system was measured by using a RAS-2 real-time aerosol photometer (MIE, Bedford, Massachusetts, USA). Samples were taken continuously from the vicinity of the breathing zone.

This chamber monitoring allows for an overall survey of toxicologically relevant technical parameters (inlet and exhaust flows as well as atmosphere homogeneity, temporal stability, and generation performance). Interruptions in exposure (e.g. resulting from obstruction of the nozzle or other technical mishaps) are recorded and, if applicable, a commensurate interval is added to the exposure duration for compensation.

7.11 Number of Animals per Group

Five male and five female rats were simultaneously exposed to each concentration under nose-only conditions for 4 h.

7.12 Control Animals

To identify exposure-related effects, comparisons with appropriate controls are conducted. Controls were exposed to aerosolized water using almost similar exposure conditions as were used for the test substance (15 liters air/min and inhalation chamber segment; duration of exposure = 1 x 4 h; 5 males and females per group). Direct comparisons were made between the control and exposure groups.

Note: Control studies are performed under GLP-conditions but without assignment to a particular study. This allows use of control data for several studies that have been performed under similar experimental conditions within a recent time frame. This procedure is in compliance with current testing guidelines as well as animal welfare regulations.

7.13 Body Weights and Duration of Observation Period

Body weights were measured before exposure, on days 3 and 7, and day 14. Individual weights are also recorded at death, if applicable. The period of observation was for 2 weeks.

7.14 Clinical Signs

The appearance and behavior of each rat were examined carefully several times on the day of exposure and at least once daily thereafter. Weekend assessments were made once a day (morning). Assessments from restraining tubes were made only if unequivocal signs occurred (e.g. spasms, abnormal movements, severe respiratory signs). Following exposure, observations are made and recorded systematically; individual records are maintained for each animal. Cage-side observations included, but were not limited to, changes in the skin and fur, eyes, mucous membranes, respiratory, circulatory, autonomic and central nervous system, and somatomotor activity and behavior pattern. Particular attention was directed to observation of tremors, convulsions, salivation, diarrhea, lethargy, somnolence and prostration. The time of death is recorded as precisely as possible, if applicable. Since these signs can only be assessed adequately from freely moving animals, no specific assessment was performed during exposure while animals were restrained.

Each rat was first observed in its home cage and then individually examined. The following reflexes were tested, based on recommendations made by Irwin (1968): visual placing response and grip strength on wire mesh, abdominal muscle tone, corneal and pupillary reflexes, pinnal reflex, righting reflex, tail-pinch response,

startle reflex with respect to behavioral changes stimulated by sounds (finger snapping) and touch (back).

All signs exceeding the exposure day are reported in the form of incidence tables (see Appendix). The resolution of these tables is per one day. The incidence of signs will reflect signs from surviving animals. Uncommon signs related to an agonal condition (if applicable) that may be observed on the day of death are recorded but may not be incorporated into the incidence tables. Those signs are described in the results section if they differ from previous observations.

7.15 Rectal Temperatures

The rectal temperatures were measured directly after cessation of exposure (approximately within ½ hour after the end of exposure) using a Digimed digital thermometer with a rectal probe for rats.

7.16 Necropsy

All surviving rats were sacrificed at the end of the observation period using sodium pentobarbital (Nembutal®) (approximately 300 mg/kg body weight, intraperitoneal injection). All rats, irrespective of the day of death, were given a gross-pathological examination. Consideration was given to performing a gross necropsy on animals as indicated by the nature of toxic effects, with particular reference to changes related to the respiratory tract. All gross pathological changes were recorded and evaluated.

7.17 Statistical Evaluation of Data

Necropsy findings: If specific findings occur from the respiratory tract of surviving rats they are evaluated statistically using the pair-wise Fisher test after the R x C chi-squared test. The Fisher test was only performed if differences occurred between groups in the R x C chi-squared test or if a frequency value of < 5 was calculated. This procedure was performed in accordance with Gad and Weil (1982). For calculation of the unilateral p value a symmetrical distribution was assumed (p unilateral = (p bilateral)/2).

Body weights: Means and single standard deviations of body weights are calculated. Mean body weights are also depicted graphically as a function of time (see result section). Since in acute studies individual group means may differ prior to commencement of the first exposure, the body weight gain was statistically

evaluated for each group. For these evaluations a one-way ANOVA (vide infra) is used.

Particle size analysis: The statistical methods used in the evaluation of the particle-size distribution were described in Section 7.8.

Physiological data: Data of rectal temperature measurements are statistically evaluated using the ANOVA procedure (vide infra).

Calculation of the LC_{50}: Due to the low number of dose groups the median lethal concentration (LC_{50}) was not calculated. However, exposure concentrations tested were used to determine the limits / ranges where a 50% probability of death can be expected.

Randomization: A computerized list of random numbers served the purpose to assign animals at random to the treatment groups.

Analysis of variance (ANOVA): This parametric method checks for normal distribution of data by comparing the median and mean. The groups are compared at a confidence level of $(1-\alpha) = 95\%$ ($p = 0.05$). The test for the between-group homogeneity of the variance employed Box's test if more than 2 study groups were compared with each other. If the above F-test shows that the intra-group variability is greater than the inter-group variability, this is shown in the Appendix as *"no statistical difference between the groups"*. If a difference is found then a pairwise post-hoc comparison is conducted (1- and 2-sided) using the Games and Howell modification of the Tukey-Kramer significance test. This program was originally obtained from BCTIC.

7.18 Programming and Validating Software

Software code for the following purposes was written in Microsoft Fortran 77: particle-size analysis, ANOVA, Fisher test, inhalation chamber and physiological data tabulation program, graphics software. The computer programs were carefully validated. The validation was conducted using published text book data sets (e.g. BCTIC, Gad and Weil, 1982). However, it should be emphasized that the formal GLP-requirements required for software validation were not fulfilled. Wherever possible, raw data and calculated/derived values are displayed graphically to provide a versatile opportunity for data comparison.

7.19 Presentation of raw data

Raw data entered into, processed and/or stored by a computer system can be saved and printed out in various formats. The precision (number of decimal places) of the figures printed out and reproduced in this report reflects the toxicologically relevant precision in all cases. Deviations between manually calculated and computer-determined figures can thus arise due to rounding. A "zero" number of decimal places does not necessarily represent the pertinent measurement precision of the detection system.

Necropsy findings: These data were presented in a 'stochiastic' manner, i.e., as being absent or present. No emphasis was made to incorporate raw data addressing subtle color changes or subjective intensity measures. Intensities or colors were only incorporated into the report when required to describe the severity of specific effects.

7.20 Archiving the Raw Data and the Report

The protocol, raw data, and the final report are archived in locations specified by Bayer AG, in accordance with GLP requirements.

8 RESULTS

8.1 Generation and Characterization of Atmosphere

Technical information concerning generation of test atmospheres is provided in Table 1.

Table 1: Generation and characterization of chamber atmosphere (liquid aerosol)

	Group 1	Group 2	Group 3
Target Conc. (mg/m³)	**0**	**750**	**2000**
Nominal Conc. (mg/m³)	333333	5862.5	11862.5
Gravimetric Conc. (mg/m³)	Control Water	690	1535
Actual concentration (mg/m³)		989 [1)]	2199 [1)]
Inlet Air Flow (l/min)	15	15	15
Dilution Air Flow (l/min)	--	7.5	--
Exhaust Air Flow (l/min)	13	13	13
Temperature (mean, °C)	21.9	21.6	21.3
Rel. Humidity (mean, %)	≈100	>95	>95
MMAD (µm)	--	2.88	4.18
GSD	--	1.66	2.06
Aerosol Mass < 3 µm (%)	--	53.4	32.4
Mass recovered (mg/m³)	--	629	1493

1) Calculated concentration of test substance (mass concentration x 100/69,8 (i.e. the test substance contains approximately 69,8% non-volatile constituents)
MMAD = Mass Median Aerodynamic Diameter
GSD = Geometric Standard Deviation
-- = not applicable

For specific information concerning calculations of aerosol MMAD, GSD, and mass dependent size fraction below 3 µm, see Appendix.

Characterization of the test atmospheres: Real-time monitoring of the aerosol test atmosphere from the breathing zone indicated that the exposure conditions were temporally stable over the exposure period.

Analysis of the aerosol particle-size distribution from the breathing zone samples demonstrates that the aerosol generated was, in general, in the respirable range. Repeated measurements made during one exposure demonstrated temporally stable particle-size distributions. The lower gravimetrical concentrations compared with the nominal concentrations are attributed to the evaporation of water. The total gravimetric concentration recovered by the cascade impactor was quite comparable to that concentration found by filter analyses. From this finding it can be concluded that interstage wall losses occurring within the low-pressure critical orifice cascade impactor or potential anisokinetic sampling errors appear to be negligible (SOT, 1992, Pauluhn *et al.*, 1996).

Temperature values in the inhalation chamber were in the range suggested by the testing guidelines. The high humidity values are related to the high amount of water in the aerosol test atmosphere.

8.2 Toxicological Results

The results obtained during and after exposures of rats for 4 h to this test substance are summarized in Table 2.

Table 2: Summary of acute inhalation toxicity - 4 hour exposure to aerosolized test substance (aerosol)

N Group /sex	Target Concentration (mg/m³)	Toxicological Result	Onset and Duration of Signs	Onset of Mortality	Rectal Temperature (°C)
1 / m	0	0 / 0 / 5	--	--	38.1
2 / m	750	0 / 5 / 5	0d - 6d	--	33.5**
3 / m	2000	0 / 5 / 5	0d - 8d	--	31.2**
1 / f	0	0 / 0 / 5	--	--	38.2
2 / f	750	0 / 5 / 5	0d - 3d	--	33.6**
3 / f	2000	4 / 5 / 5	0d -14d	1d - 6d	30.2**

N = group assignment, m = males, f = females, 0d = day of exposure, **=p<0.01

Values given in the *'Toxicological results'* column are as follows:

1st number = number of dead animals
2nd number = number of animals with signs after exposure cessation
3rd number = number of animals exposed

Mortality

LC_{50}: rat, male > 2199 mg/m³

rat, female > 989 mg/m³ - < 2199 mg/m³

Different mortality figures were obtained in males and females. Mortality occured only in females of the high-dose group from day 1 after exposure until day 6 of the follow-up period.

Signs and observations

Details concerning signs and observations are provided in the Appendix in the form of various incidence tables. The following list of signs is focusing on toxicologically significant signs only.

Control Groups : All rats tolerated the exposure without specific signs.

Treatment Groups : Bradypnea, labored breathing pattern, dyspnoea, nostrils reddened and encrusted, reduced motility, atony, tremor, piloerection, salivation, ungroomed hair-coat, pupils dilated, palpebral narrowing, pallor, stilted gait, increased muscle tone, body weight loss, lying in a flattened position.

Reflex measurements

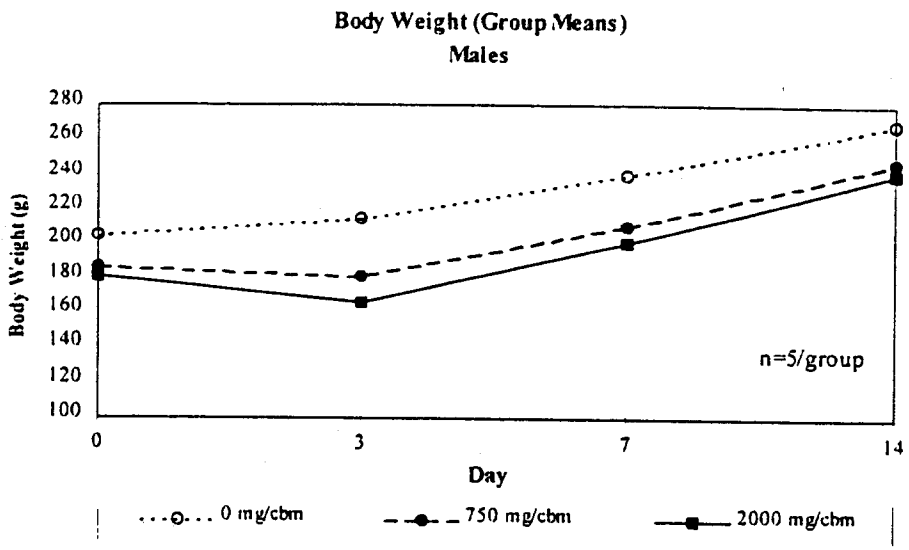
A battery of reflex measurements was made on the first postexposure day. Rats of the low-dose exposure groups of both sexes tolerated the exposure without any specific effect. High-dose females - and to a lesser extent - high-dose males exhibited abnormal reflexes which can be summarized as following:

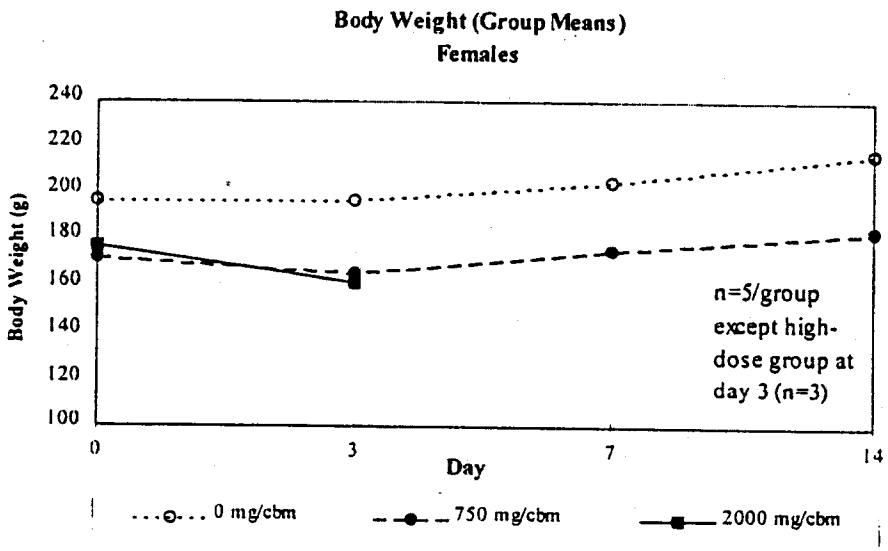
Reduced grip strength, lacking grip strength (females only), reduced muscle tone, increased muscle tone (females only), reduced pupil response to pen-light, abnormal pinna reflex (females only), abnormal startle reflex to sound and touch (females only), changes in righting responses (for details see Appendix).

Body weights

Results of the evaluation of the body weights are included in the Appendix. Due to differences in baseline means between control animals and animals exposed to the test substance, confirmative statistics were done only on body weight gains. Comparisons between control animals with those in the group exposed to the test substance revealed a statistically significant body weight loss in the exposure groups of both sexes at day 3 (except high-dose females where test statistics were not calculated due to the reduced number of animals available at that time). Statistically significant higher body weight gains occurred in males at day 7 and day 14, being interpreted as a compensatory effect following the initial phase of body weight loss. The overall change of mean body weights is depicted in Fig. 4.

Figure 4: Body Weights (means)

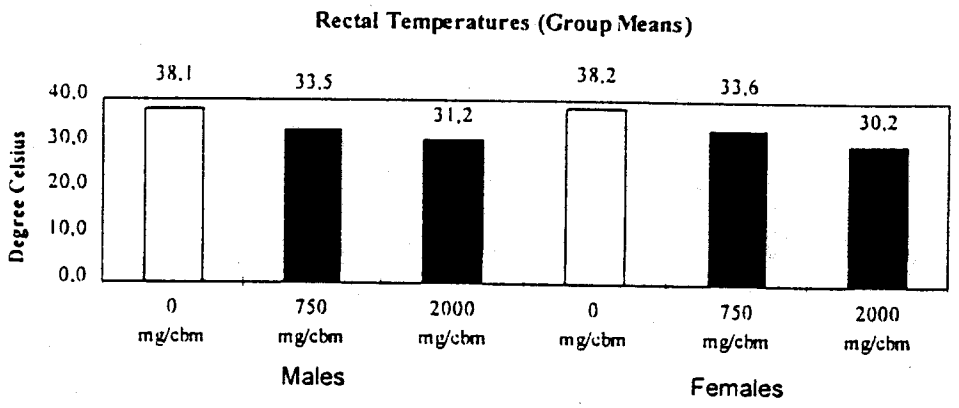




Rectal temperature

Results of the evaluation of the rectal temperature are summarised in the Appendix. Rats exposed to the test substance experienced a marked dose-dependent decrease in body temperature. Statistical comparison indicates a significantly decreased body temperature in males and females of both dose groups.

Figure 5: Rectal Temperatures (means)



Necropsy

Individual findings from the gross-pathological examinations - including the exact onset of mortality - are summarized in the Appendix. A qualitative description, only of findings of toxicological importance and for toxicological evaluation, is given below.

Animals sacrificed at the end of the observation period: In all animals exposed to the test compound and surviving the entire test period a conclusive, concentration-dependent increased incidence of macroscopic findings could not be ascertained. The discolorations of the lungs observed singularily are not considered to be causally related to the exposure of the test substance.

Animals that died intercurrently: Lungs less collapsed, dark-red discolorations of lungs, discolorations of other parenchymatous organs.

9 EVALUATION AND DISCUSSION

Exposure to the maximum tested concentration of 2199 mg/m³ was accompanied with mortality in female animals and - in both sexes - with clinical signs indicative of respiratory tract irritation (bradypnea, labored breathing pattern, dyspnoea, nostrils reddened and encrusted), hypothermia, piloerection, salivation, ungroomed hair-coat, pallor, stilted gait, increased muscle tone, palpebral narrowing, retardation in body weight gain and central nervous effects (tremor, reduced motility, atony, dilated pupils). The duration of signs appeared to be governed by respiratory signs such as laboured breathing (until day 13) and unspecific signs such as piloerection (until day 7).

A concentration of 989 mg/m³ was tolerated without mortality in both sexes but with few symptoms, mainly due to respiratory tract irritation (bradypnea, labored breathing pattern), retardation in body weight gain and central nervous effects (reduced motility and tremor). No treatment-related findings were detected in the rats sacrificed at the end of the 2-week postexposure period or found dead.

With regard to the respirability of the aerosol generated internationally recognized recommendations such as of SOT (1992) were almost fulfilled, i.e. the MMAD was less than 4 µm in the low-dose groups (MMAD ≈ 2,88 µm, GSD ≈ 1.66) but slightly above 4 µm in the high-dose group (MMAD ≈ 4,18 µm, GSD ≈ 2,06).

Inhalation of respiratory irritants is known to induce reflex changes in the breathing pattern and cardiac output and are reported to be associated with the decline in the metabolic rate and body temperature of rodents (Jaeger and Gearhart, 1982; Mautz and Bufalino, 1989). The present observation in rats may reflect a more labile thermoregulatory physiology among rodents and may involve a complex set of physiological responses that may differ among mammalian species. Sensory irritation induces respiratory and concomitantly central distress in animals which appears to be the possible cause of hypothermia.

In summary, the aerosolized test substance (liquid aerosol) proved to have a moderate acute inhalation toxicity to female rats and a low acute inhalation toxicity to

male rats. Specific pathognomonic findings indicate a causal relationship between exposure concentration and signs related to respiratory tract irritation.

10 KEY TO ABBREVIATIONS

Konz.	Concentration
nomin.	Nominal
analyt.	Analytical
mcm/µm	Micrometer
Expos.	Exposure
MMAD	Mass Median Aerodynamic Diameter
NMAD	Number Median Aerodynamic Diameter
GSD	Geometric standard deviation (GSD)
ECD	Effective cut-off diameter
Ai	Sample for analysis
STAND, S, Std, s	Standard deviation
MW/MEANS	Means
F	F-test value (F-ratio)
DF	Degrees of freedom
PROB	Probability
SS	Total sum of squares
MS	Mean squares
TREATMENT	- between the groups
ERROR	- within the groups
TOTAL	- total

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BCTIC Computer Code Collection - Biomedical computing Technology Information Center, ANOVA a Fortran Program to Perform one-way Classification Analysis of Variance. Vanderbilt Medical Center, Nashville Tennessee, U.S.A.

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12 APPENDIX

Atmosphere Characterization

Group	Date of exposure (DD.MM.YY)	Sampled Volume (l) / flow rate (l/min)	Target Conc. (mg/m³ air)	Actual Concentrations (mg/m³ air)	Mean Conc. (mg/m³ air)
1	10.08.1998	n.a.	--	0 (control)	n.a.
3	16.09.1998	10 / 4 **(filter analysis)**	750	670 690 740 660	690 (989) [1)]
2	14.09.1998	10 / 4 **(filter analysis)**	2000	1470 1460 1630 1580	1535 (2199) [1)]

1) Calculated concentration of test substance: mass concentration x 100/69,8 (i.e. the test substance contains approximately 69,8% non-volatile constituents)

In the subsequently presented tables, in most instances, target concentrations are used to indicate the respective group.

Characterization of Particle Size Distribution

ANALYSIS OF PARTICLE DISTRIBUTIONS

Type of investigation: Acute Inhalation - Aerosol
Compound: YRC 2894 480 SC 05776/0096 (I)
Date of exposure: 14.09.98 Study-no.: T6067418
Target concentration: 2000.0 mg/m3 air

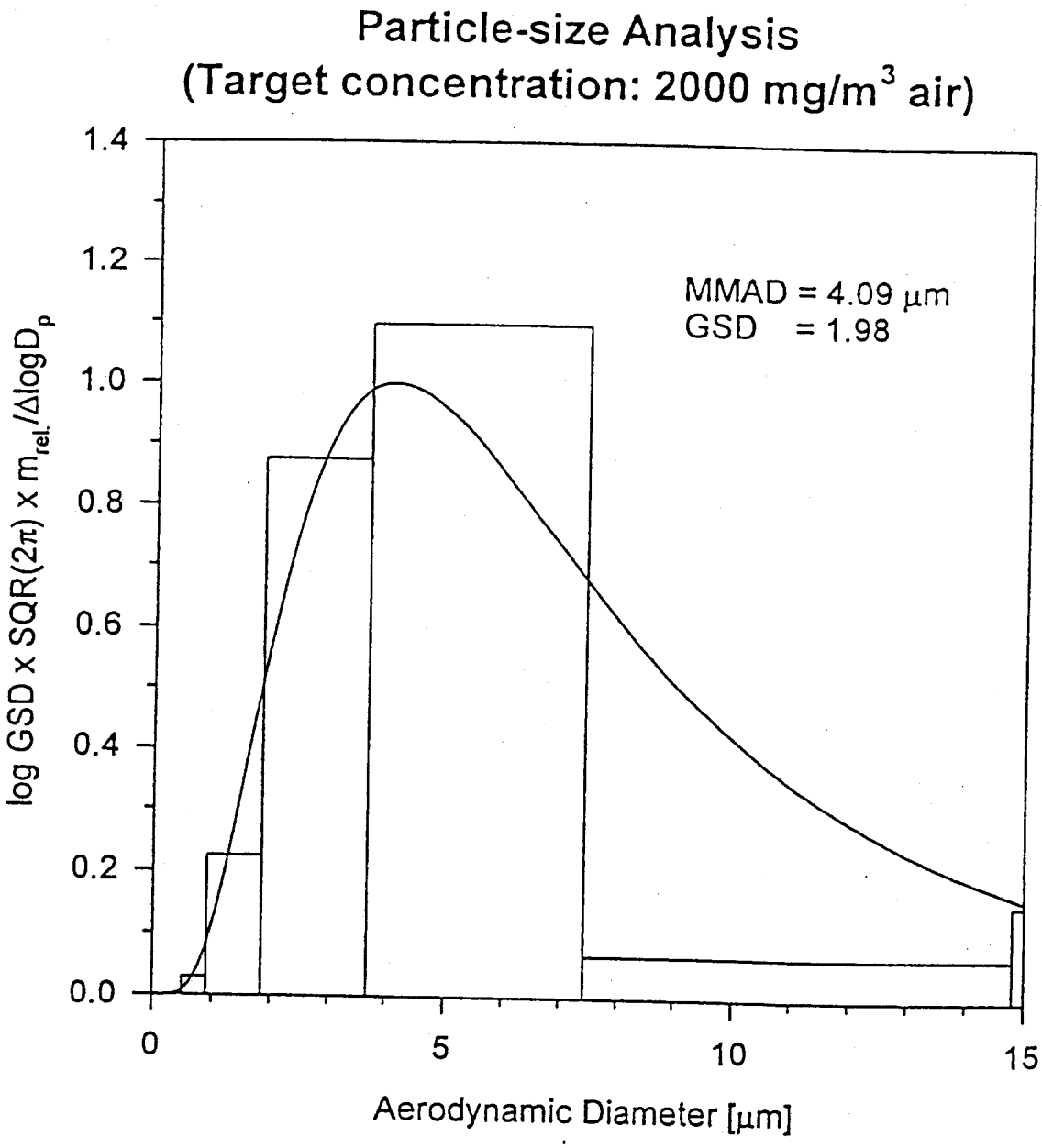
N	Impactor stage (µm - µm)	Cut-Off diameter (µm)	Mass/ stage (mg)	Rel. mass (%)	Cumul. mass (%)
1	0.06 - 0.12	0.060	0.000	0.00	0.00
2	0.12 - 0.25	0.120	0.001	0.01	0.00
3	0.25 - 0.49	0.250	0.008	0.05	0.01
4	0.49 - 0.90	0.490	0.187	1.08	0.05
5	0.90 - 1.85	0.900	1.642	9.47	1.13
6	1.85 - 3.69	1.850	6.144	35.43	10.60
7	3.69 - 7.42	3.690	7.785	44.89	46.03
8	7.42 -14.80	7.420	0.474	2.73	90.92
9	14.80 -30.00	14.800	1.100	6.34	93.66

Mass Median Aerodynamic Diameter (MMAD): 4.09 µm
Geometric standard deviation (GSD): 1.98
Number Median Aerodynamic Diameter (NMAD): 1.02 µm
Surface Median Aerodynamic Diameter (SMAD): 2.57 µm
System: BERNER-IMPACTOR I
Air flow: 5.85 liter/min.
Sampling time: 120.00 seconds
Concentration (computed): 1482.14 mg/m3 air

Respirability (percent < 1.0 µm):
1. Mass related: 2.1 % (measured)
2. Number related: 49.1 % (extrapolated)

Respirability (percent < 3.0 µm):
1. Mass related: 32.7 % (measured)
2. Number related: 94.5 % (extrapolated)

Respirability (percent < 5.0 µm):
1. Mass related: 61.7 % (measured)
2. Number related: 99.1 % (extrapolated)


Particle-size Analysis
(Target concentration: 2000 mg/m3 air)
MMAD = 4.09 µm
GSD = 1.98
log GSD x SQR(2π) x m_rel./ΔlogD_p
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0
5
10
15
Aerodynamic Diameter [µm]

ANALYSIS OF PARTICLE DISTRIBUTIONS

Type of investigation: Acute Inhalation - Aerosol
Compound: YRC 2894 480 SC 05776/0096 (II)
Date of exposure: 14.09.98 Study-no.: T6067418
Target concentration: 2000.0 mg/m3 air

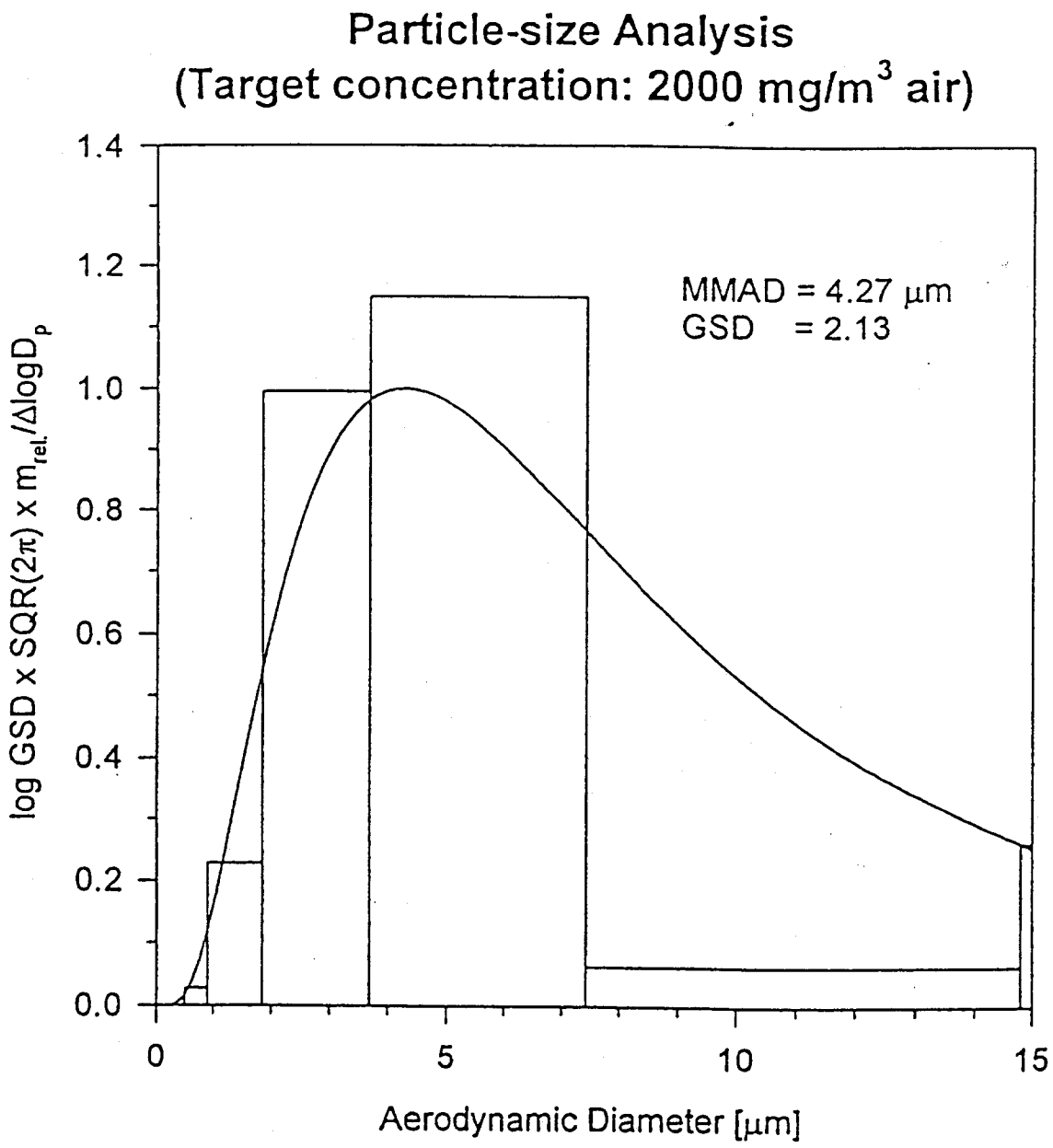
N	Impactor stage (µm - µm)	Cut-Off diameter (µm)	Mass/ stage (mg)	Rel. mass (%)	Cumul. mass (%)
1	0.06 - 0.12	0.060	0.000	0.00	0.00
2	0.12 - 0.25	0.120	0.000	0.00	0.00
3	0.25 - 0.49	0.250	0.000	0.00	0.00
4	0.49 - 0.90	0.490	0.158	0.90	0.00
5	0.90 - 1.85	0.900	1.524	8.66	0.90
6	1.85 - 3.69	1.850	6.369	36.20	9.56
7	3.69 - 7.42	3.690	7.436	42.27	45.76
8	7.42 -14.80	7.420	0.406	2.31	88.03
9	14.80 -30.00	14.800	1.700	9.66	90.34

Mass Median Aerodynamic Diameter (MMAD): 4.27 µm
Geometric standard deviation (GSD): 2.13
Number Median Aerodynamic Diameter (NMAD): 0.77 µm
Surface Median Aerodynamic Diameter (SMAD): 2.41 µm
System: BERNER-IMPACTOR I
Air flow: 5.85 liter/min.
Sampling time: 120.00 seconds
Concentration (computed): 1503.68 mg/m3 air

Respirability (percent < 1.0 µm):
1. Mass related: 2.9 % (measured)
2. Number related: 63.9 % (extrapolated)

Respirability (percent < 3.0 µm):
1. Mass related: 32.1 % (measured)
2. Number related: 96.5 % (extrapolated)

Respirability (percent < 5.0 µm):
1. Mass related: 58.3 % (measured)
2. Number related: 99.1 % (extrapolated)


Particle-size Analysis
(Target concentration: 2000 mg/m3 air)
MMAD = 4.27 µm
GSD = 2.13
log GSD x SQR(2π) x m_rel./ΔlogD_p
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0
5
10
15
Aerodynamic Diameter [µm]

ANALYSIS OF PARTICLE DISTRIBUTIONS

Type of investigation: Acute Inhalation - Aerosol
Compound: YRC 2894 480 SC 05776/0096 (I)
Date of exposure: 16.09.98 Study-no.: T6067418
Target concentration: 750.0 mg/m3 air

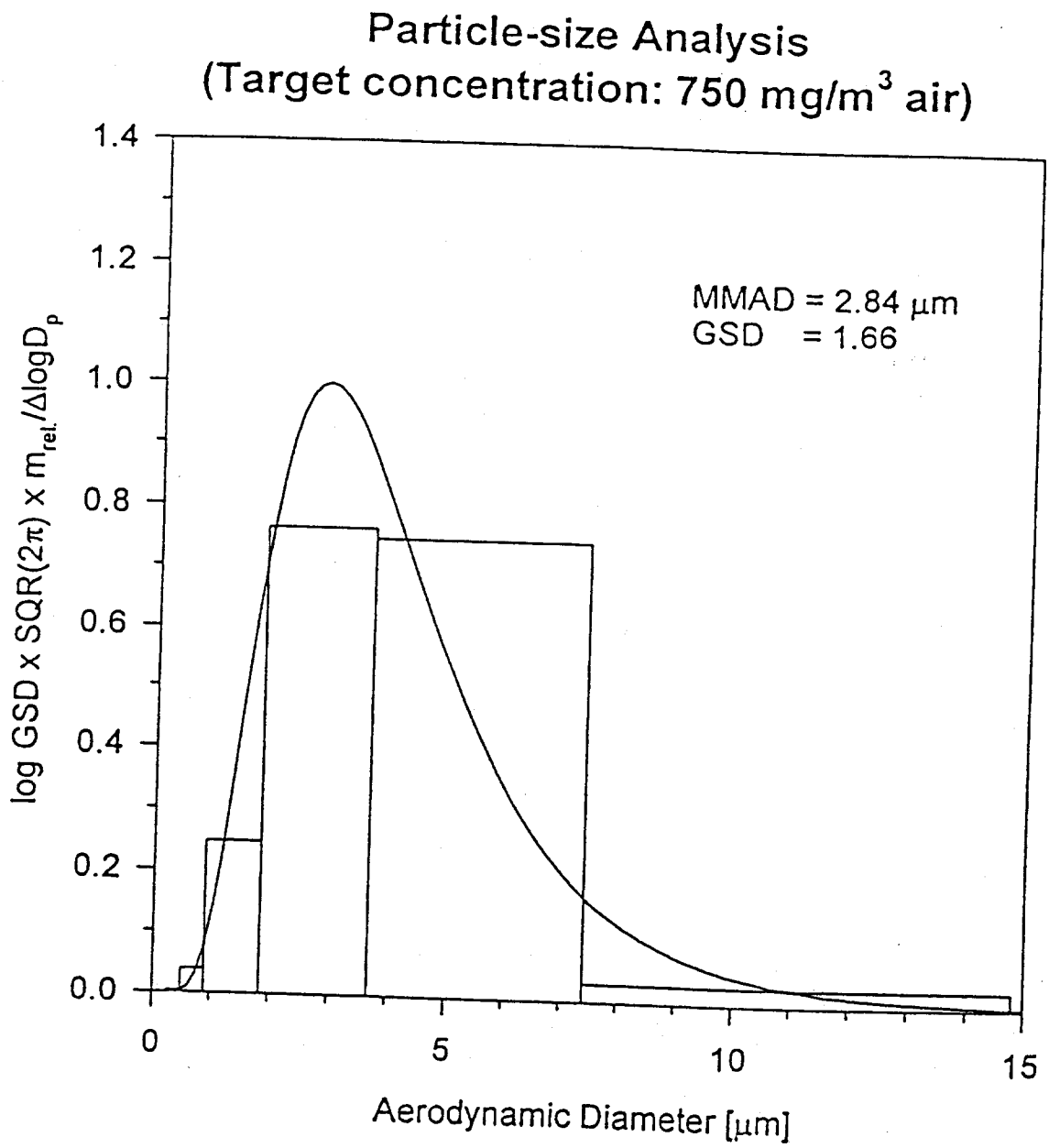
N	Impactor stage (µm - µm)	Cut-Off diameter (µm)	Mass/ stage (mg)	Rel. mass (%)	Cumul. mass (%)
1	0.06 - 0.12	0.060	0.000	0.00	0.00
2	0.12 - 0.25	0.120	0.002	0.01	0.00
3	0.25 - 0.49	0.250	0.028	0.20	0.01
4	0.49 - 0.90	0.490	0.266	1.85	0.21
5	0.90 - 1.85	0.900	2.003	13.96	2.06
6	1.85 - 3.69	1.850	5.942	41.42	16.03
7	3.69 - 7.42	3.690	5.871	40.93	57.45
8	7.42 -14.80	7.420	0.232	1.62	98.38
9	14.80 -30.00	14.800	0.000	0.00	100.00

Mass Median Aerodynamic Diameter (MMAD): 2.84 µm
Geometric standard deviation (GSD): 1.66
Number Median Aerodynamic Diameter (NMAD): 1.31 µm
Surface Median Aerodynamic Diameter (SMAD): 2.20 µm
System: BERNER-IMPACTOR I
Air flow: 5.85 liter/min.
Sampling time: 240.00 seconds
Concentration (computed): 612.99 mg/m3 air

Respirability (percent < 1.0 µm):
1. Mass related: 2.1 % (measured)
2. Number related: 30.1 % (extrapolated)

Respirability (percent < 3.0 µm):
1. Mass related: 54.3 % (measured)
2. Number related: 94.9 % (extrapolated)

Respirability (percent < 5.0 µm):
1. Mass related: 86.5 % (measured)
2. Number related: 99.1 % (extrapolated)


Particle-size Analysis
(Target concentration: 750 mg/m3 air)
MMAD = 2.84 µm
GSD = 1.66
log GSD x SQR(2π) x m_rel./ΔlogD_p
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0
5
10
15
Aerodynamic Diameter [µm]

ANALYSIS OF PARTICLE DISTRIBUTIONS

Type of investigation: Acute Inhalation - Aerosol
Compound: YRC 2894 480 SC 05776/0096 (II)
Date of exposure: 16.09.98 Study-no.: T6067418
Target concentration 750.0 mg/m3 air

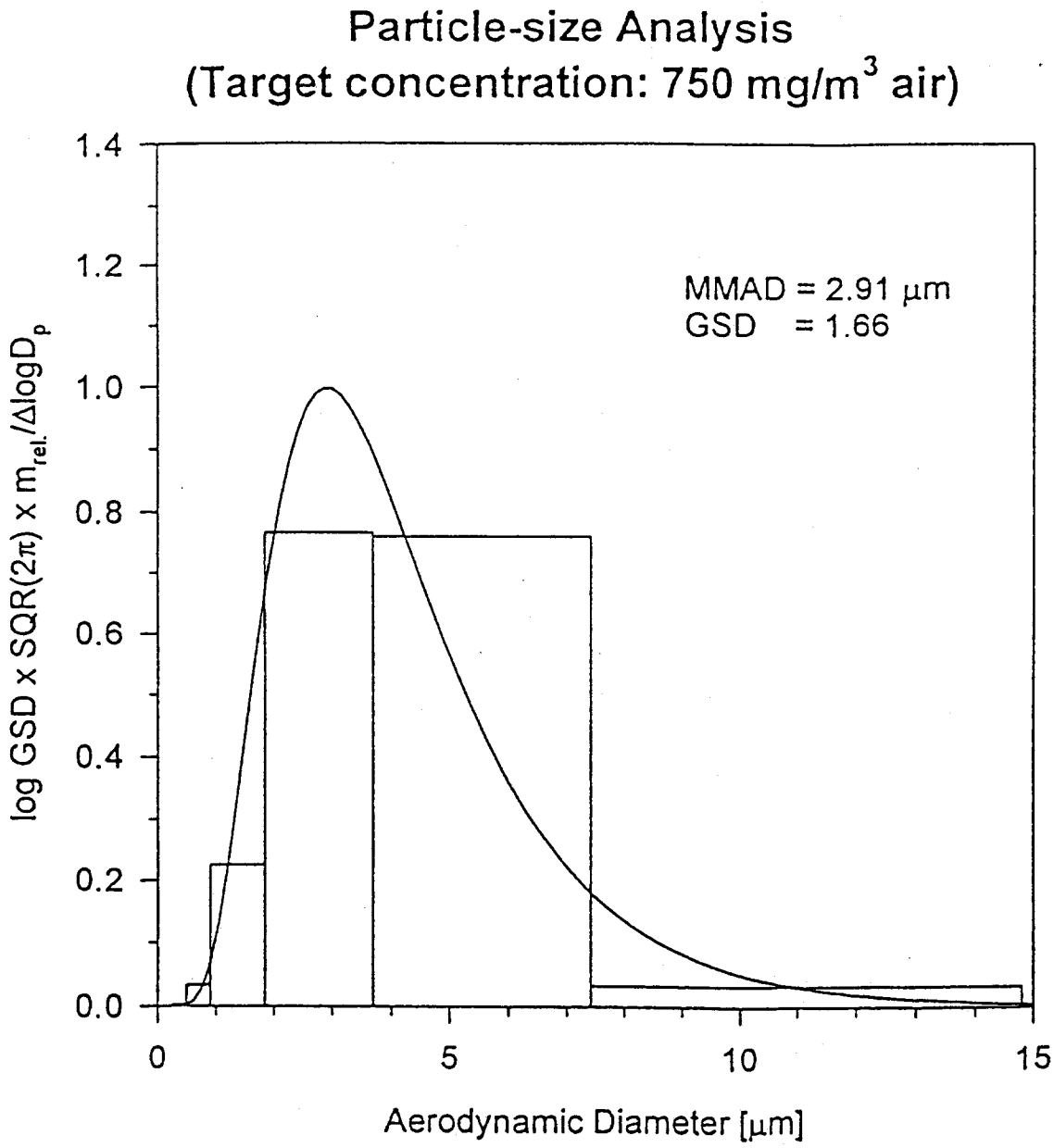
N	Impactor stage (µm - µm)	Cut-Off diameter (µm)	Mass/ stage (mg)	Rel. mass (%)	Cumul. mass (%)
1	0.06 - 0.12	0.060	0.000	0.00	0.00
2	0.12 - 0.25	0.120	0.000	0.00	0.00
3	0.25 - 0.49	0.250	0.029	0.19	0.00
4	0.49 - 0.90	0.490	0.245	1.62	0.19
5	0.90 - 1.85	0.900	1.931	12.79	1.82
6	1.85 - 3.69	1.850	6.295	41.71	14.61
7	3.69 - 7.42	3.690	6.323	41.90	56.32
8	7.42 - 14.80	7.420	0.269	1.78	98.22
9	14.80 - 30.00	14.800	0.000	0.00	100.00

Mass Median Aerodynamic Diameter (MMAD): 2.91 µm
Geometric standard deviation (GSD): 1.66
Number Median Aerodynamic Diameter (NMAD): 1.35 µm
Surface Median Aerodynamic Diameter (SMAD): 2.25 µm
System: BERNER-IMPACTOR I
Air flow: 5.85 liter/min.
Sampling time: 240.00 seconds
Concentration (computed): 644.96 mg/m3 air

Respirability (percent < 1.0 µm):
1. Mass related: 1.9 % (measured)
2. Number related: 27.9 % (extrapolated)

Respirability (percent < 3.0 µm):
1. Mass related: 52.5 % (measured)
2. Number related: 94.3 % (extrapolated)

Respirability (percent < 5.0 µm):
1. Mass related: 85.7 % (measured)
2. Number related: 99.1 % (extrapolated)


Particle-size Analysis
(Target concentration: 750 mg/m3 air)
MMAD = 2.91 µm
GSD = 1.66
log GSD x SQR(2π) x m_rel./ΔlogD_p
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0
5
10
15
Aerodynamic Diameter [µm]

Reflexes

Measurements on day: 1 / MALES

Type of Reflex	Group 1	Group 2	Group 3
Target Concentration (mg/m^3)	**0**	**750**	**2000**
Number of animals investigated	**5**	**5**	**5**
Visual placing response	0	0	0
Grip strength (vertical)	0	0	2[a]
Grip strength (horizontal)	0	0	2[a]
Tonus	0	0	1[a]
Cornea reflex	0	0	0
Light reflex	0	0	2[a]
Pinna reflex	0	0	0
Startle reflex / sound	0	0	0
Startle reflex / touch	0	0	0
Tail-pinch response	0	0	0
Righting response (open field)	0	0	0
Righting response (drop method)	0	0	2[b]

[a] : reduced [b] : slightly uncoordinated

#: number of rats showing abnormal reflexes

Measurements on day:1 / FEMALES

Type of Reflex	Group 1	Group 2	Group 3
Target Concentration (mg/m^3)	0	750	2000
Number of animals investigated	5	5	4
Visual placing response	0	0	1
Grip strength (vertical)	0	0	2[a]
Grip strength (horizontal)	0	0	2[b]
Tonus	0	0	3[c]
Cornea reflex	0	0	0
Light reflex	0	0	3[d]
Pinna reflex	0	0	1
Startle reflex / sound	0	0	1[e]
Startle reflex / touch	0	0	1[f]
Tail-pinch response	0	0	0
Righting response (open field)	0	0	4[g]
Righting response (drop method)	0	0	4[h]

[a] : *one*: reduced *one*: absent
[b] : *one*: reduced *one*: absent
[c] : *two*: increased *one*: reduced
[d] : *two*: mydriasis *one*: miosis
[e] : absent
[f] : freezing
[g] : *three*: impaired *one*: remains on back
[h] : *three*: slightly uncoordinated *one*: lands on side

#: number of rats showing abnormal reflexes

Rectal Temperatures

Rectal temperature (° C): Male Animals

Control Animal no.	1	2	3	4	5
	37.8	38.1	38.4	37.9	38.2
750 mg/cbm Animal no.	21	22	23	24	25
	33.8	33.5	35.4	32.3	32.6
2000 mg/cbm Animal no.	11	12	13	14	15
	30.2	30.3	31.2	29.9	34.2

Rectal temperature (° C): Female Animals

Control Animal no.	6	7	8	9	10
	38.4	38.1	38.1	38.0	38.2
750 mg/cbm Animal no.	26	27	28	29	30
	33.2	33.2	32.9	35.6	33.3
2000 mg/cbm Animal no.	16	17	18	19	20
	30.3	29.7	29.8	29.9	31.4

YRC 2894 480 SC 05776/0096 T6067418

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Rect.Temp (Deg.Cel) / MALES

Group-no.: 1 (1-5)

37.800	38.100	38.400	37.900	38.200

MEDIAN= 38.100 MEAN= 38.080 STD = .239

Group-no.: 2 (21-25)

33.800	33.500	35.400	32.300	32.600

MEDIAN= 33.500 MEAN= 33.520 STD = 1.219

Group-no.: 3 (11-15)

30.200	30.300	31.200	29.900	34.200

MEDIAN= 30.300 MEAN= 31.160 STD = 1.767

BOXs TEST FOR HOMOGENEITY OF VARIANCES AT P=.05000 LEVEL

CALCULATED F	D.F.s	PROBABILITY
4.8962	2 & 324.	.0082

HETEROGENEOUS VARIANCES (ONE-TAILED TEST)

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	123.7	2	61.875	39.774	.000
ERROR	18.67	12	1.5557		
TOTAL	142.4	14			

OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL

GAMES AND HOWELL MODIFICATION OF
TUKEY-KRAMERs HONESTLY SIGNIFICANT DIFFERENCE TEST
(WITH THE STUDENTIZED RANGE STATISTIC)

GROUPS COMPARED	CALCULATED TEST VALUE	DEGREES OF FREEDOM	PROBABILITY	CONCLUSION
5. % ONE-TAILED TEST				
1 AND 2	-11.60	4	.0013	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 2	11.60	4	.0013	SIGNIFICANT
5. % ONE-TAILED TEST				
1 AND 3	-12.27	4	.0007	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 3	12.27	4	.0007	SIGNIFICANT
5. % ONE-TAILED TEST				
2 AND 3	-3.48	7	.0976	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
2 AND 3	3.48	7	.0976	NOT SIGNIFICANT

YRC 2894 480 SC 05776/0096 T6067418

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Rect.Temp (Deg.Cel) / FEMALES

Group-no.: 1 (6-10)

38.400	38.100	38.100	38.000	38.200

MEDIAN= 38.100 MEAN= 38.160 STD = .152

Group-no.: 2 (26-30)

33.200	33.200	32.900	35.600	33.300

MEDIAN= 33.200 MEAN= 33.640 STD = 1.106

Group-no.: 3 (16-20)

30.300	29.700	29.800	29.900	31.400

MEDIAN= 29.900 MEAN= 30.220 STD = .698

BOXs TEST FOR HOMOGENEITY OF VARIANCES AT P=.05000 LEVEL

CALCULATED F	D.F.s	PROBABILITY
4.8759	2 & 324.	.0084

HETEROGENEOUS VARIANCES (ONE-TAILED TEST)

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	158.6	2	79.309	137.291	.000
ERROR	6.932	12	.57767		
TOTAL	165.5	14			

OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL

GAMES AND HOWELL MODIFICATION OF
TUKEY-KRAMERs HONESTLY SIGNIFICANT DIFFERENCE TEST
(WITH THE STUDENTIZED RANGE STATISTIC)

GROUPS COMPARED	CALCULATED TEST VALUE	DEGREES OF FREEDOM	PROBABILITY	CONCLUSION
5. % ONE-TAILED TEST				
1 AND 2	-12.80	4	.0005	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 2	12.80	4	.0005	SIGNIFICANT
5. % ONE-TAILED TEST				
1 AND 3	-35.16	4	.0000	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 3	35.16	4	.0000	SIGNIFICANT
5. % ONE-TAILED TEST				
2 AND 3	-8.27	7	.0016	SIGNIFICANT
5. % TWO-TAILED TEST				
2 AND 3	8.27	7	.0016	SIGNIFICANT

Body Weights

Analysis of Body Weights [all data in g]

Group 1: Control - MALES

	Postexposure Day			
	0	3	7	14
1	202.	212.	241.	274.
2	203.	214.	234.	264.
3	209.	218.	246.	273.
4	209.	217.	238.	267.
5	203.	214.	241.	270.
MEAN	205.2	215.0	240.0	269.6
STD	3.5	2.4	4.4	4.2

Group 2: 750 mg/m^3 - MALES

	Postexposure Day			
	0	3	7	14
21	188.	180.	209.	245.
22	195.	194.	225.	265.
23	187.	180.	210.	252.
24	181.	171.	201.	230.
25	182.	177.	206.	243.
MEAN	186.6	180.4	210.2	247.0
STD	5.6	8.4	9.0	12.8

Group 3: 2000 mg/m^3 - MALES

	Postexposure Day			
	0	3	7	14
11	184.	162.	199.	247.
12	184.	183.	210.	245.
13	179.	165.	203.	243.
14	175.	151.	186.	225.
15	182.	166.	203.	242.
MEAN	180.8	165.4	200.2	240.4
STD	3.8	11.5	8.9	8.8

Analysis of Body Weights [all data in g]

Group 1: Control - FEMALES

	Postexposure Day			
	0	3	7	14
6	203.	201.	210.	224.
7	190.	194.	204.	214.
8	195.	197.	203.	214.
9	200.	195.	203.	218.
10	193.	195.	204.	214.
MEAN	196.2	196.4	204.8	216.8
STD	5.3	2.8	2.9	4.4

Group 2: 750 mg/m^3 - FEMALES

	Postexposure Day			
	0	3	7	14
26	169.	164.	175.	189.
27	167.	159.	168.	176.
28	173.	166.	179.	186.
29	171.	162.	172.	183.
30	182.	176.	182.	182.
MEAN	172.4	165.4	175.2	183.2
STD	5.8	6.5	5.5	4.9

Group 3: 2000 mg/m^3 - FEMALES

	Postexposure Day			
	0	3	7	14
16	168.			
17	194.	169.		
18	175.	147.		
19	181.	169.	177.	193.
20	165.			
MEAN	176.6	161.7		
STD	11.5	12.7		

Analysis of Body Weight Gains [all data in g]

Group 1: Control - MALES

	Postexposure Day 3	7	14
1	10.00	29.00	33.00
2	11.00	20.00	30.00
3	9.00	28.00	27.00
4	8.00	21.00	29.00
5	11.00	27.00	29.00
MEAN	9.8	25.0	29.6
STD	1.3	4.2	2.2

Group 2: 750 mg/m^3 - MALES

	Postexposure Day 3	7	14
21	-8.00	29.00	36.00
22	-1.00	31.00	40.00
23	-7.00	30.00	42.00
24	-10.00	30.00	29.00
25	-5.00	29.00	37.00
MEAN	-6.2	29.8	36.8
STD	3.4	.8	5.0

Group 3: 2000 mg/m^3 - MALES

	Postexposure Day		
	3	7	14
11	-22.00	37.00	48.00
12	-1.00	27.00	35.00
13	-14.00	38.00	40.00
14	-24.00	35.00	39.00
15	-16.00	37.00	39.00
MEAN	-15.4	34.8	40.2
STD	9.0	4.5	4.8

Analysis of Body Weight Gains [all data in g]

Group 1: Control - FEMALES

	Postexposure Day		
	3	7	14
6	-2.00	9.00	14.00
7	4.00	10.00	10.00
8	2.00	6.00	11.00
9	-5.00	8.00	15.00
10	2.00	9.00	10.00
MEAN	.2	8.4	12.0
STD	3.6	1.5	2.3

Group 2: 750 mg/m^3 - FEMALES

	Postexposure Day		
	3	7	14
26	-5.00	11.00	14.00
27	-8.00	9.00	8.00
28	-7.00	13.00	7.00
29	-9.00	10.00	11.00
30	-6.00	6.00	.00
MEAN	-7.0	9.8	8.0
STD	1.6	2.6	5.2

Group 3: 2000 mg/m^3 - FEMALES

	Postexposure Day		
	3	7	14
16			
17	-25.00		
18	-28.00		
19	-12.00	8.00	16.00
20			
MEAN	-21.7		
STD	8.5		

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Day: 3 / MALES

Group-no.: 1

10.000	11.000	9.000	8.000	11.000

MEDIAN= 10.000 MEAN= 9.800 STD = 1.304

Group-no.: 2

-8.000	-1.000	-7.000	-10.000	-5.000

MEDIAN= -7.000 MEAN= -6.200 STD = 3.421

Group-no.: 3

-22.000	-1.000	-14.000	-24.000	-16.000

MEDIAN= -16.000 MEAN= -15.400 STD = 9.044

BOXs TEST FOR HOMOGENEITY OF VARIANCES AT P=.05000 LEVEL

CALCULATED F	D.F.s	PROBABILITY
5.5116	2 & 324.	.0048

HETEROGENEOUS VARIANCES (ONE-TAILED TEST)

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	1626.	2	813.07	25.622	.000
ERROR	380.8	12	31.733		
TOTAL	2007.	14			

OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL

GAMES AND HOWELL MODIFICATION OF
TUKEY-KRAMERs HONESTLY SIGNIFICANT DIFFERENCE TEST
(WITH THE STUDENTIZED RANGE STATISTIC)

GROUPS COMPARED	CALCULATED TEST VALUE	DEGREES OF FREEDOM	PROBABILITY	CONCLUSION
5. % ONE-TAILED TEST				
1 AND 2	-13.82	5	.0001	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 2	13.82	5	.0001	SIGNIFICANT
5. % ONE-TAILED TEST				
1 AND 3	-8.72	4	.0074	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 3	8.72	4	.0074	SIGNIFICANT
5. % ONE-TAILED TEST				
2 AND 3	-3.01	5	.1786	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
2 AND 3	3.01	5	.1786	NOT SIGNIFICANT

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Day: 7 / MALES

Group-no.: 1

29.000	20.000	28.000	21.000	27.000
MEDIAN= 27.000	MEAN= 25.000	STD = 4.183		

Group-no.: 2

29.000	31.000	30.000	30.000	29.000
MEDIAN= 30.000	MEAN= 29.800	STD = .837		

Group-no.: 3

37.000	27.000	38.000	35.000	37.000
MEDIAN= 37.000	MEAN= 34.800	STD = 4.494		

BOXs TEST FOR HOMOGENEITY OF VARIANCES AT P=.05000 LEVEL

CALCULATED F	D.F.s	PROBABILITY
3.9183	2 & 324.	.0203

HETEROGENEOUS VARIANCES (ONE-TAILED TEST)

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	240.1	2	120.07	9.380	.004
ERROR	153.6	12	12.800		
TOTAL	393.7	14			

OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL

GAMES AND HOWELL MODIFICATION OF
TUKEY-KRAMERs HONESTLY SIGNIFICANT DIFFERENCE TEST
(WITH THE STUDENTIZED RANGE STATISTIC)

GROUPS COMPARED	CALCULATED TEST VALUE	DEGREES OF FREEDOM	PROBABILITY	CONCLUSION
5. % ONE-TAILED TEST				
1 AND 2	3.56	4	.1339	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 2	3.56	4	.1339	NOT SIGNIFICANT
5. % ONE-TAILED TEST				
1 AND 3	5.05	8	.0178	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 3	5.05	8	.0178	SIGNIFICANT
5. % ONE-TAILED TEST				
2 AND 3	3.46	4	.1437	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
2 AND 3	3.46	4	.1437	NOT SIGNIFICANT

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Day: 14 / MALES

Group-no.: 1

33.000	30.000	27.000	29.000	29.000
MEDIAN= 29.000	MEAN= 29.600	STD = 2.191		

Group-no.: 2

36.000	40.000	42.000	29.000	37.000
MEDIAN= 37.000	MEAN= 36.800	STD = 4.970		

Group-no.: 3

48.000	35.000	40.000	39.000	39.000
MEDIAN= 39.000	MEAN= 40.200	STD = 4.764		

BOXs TEST FOR HOMOGENEITY OF VARIANCES AT P=.05000 LEVEL

CALCULATED F	D.F.s	PROBABILITY
1.2113	2 & 324.	.2988

HOMOGENEOUS VARIANCES (ONE-TAILED TEST)

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	292.9	2	146.47	8.418	.005
ERROR	208.8	12	17.400		
TOTAL	501.7	14			

OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL

GAMES AND HOWELL MODIFICATION OF
TUKEY-KRAMERs HONESTLY SIGNIFICANT DIFFERENCE TEST
(WITH THE STUDENTIZED RANGE STATISTIC)

GROUPS COMPARED	CALCULATED TEST VALUE	DEGREES OF FREEDOM	PROBABILITY	CONCLUSION
5. % ONE-TAILED TEST				
1 AND 2	4.19	5	.0685	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 2	4.19	5	.0685	NOT SIGNIFICANT
5. % ONE-TAILED TEST				
1 AND 3	6.39	6	.0096	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 3	6.39	6	.0096	SIGNIFICANT
5. % ONE-TAILED TEST				
2 AND 3	1.56	8	.5380	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
2 AND 3	1.56	8	.5380	NOT SIGNIFICANT

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Day: 3 / FEMALES

Group-no.: 1

-2.000 4.000 2.000 -5.000 2.000

MEDIAN= 2.000 MEAN= .200 STD = 3.633

Group-no.: 2

-5.000 -8.000 -7.000 -9.000 -6.000

MEDIAN= -7.000 MEAN= -7.000 STD = 1.581

Group-no.: 3

-25.000 -28.000 -12.000

MEDIAN= -25.000 MEAN= -21.667 STD = 8.505

BOXs TEST FOR HOMOGENEITY OF VARIANCES AT P=.05000 LEVEL

CALCULATED F	D.F.s	PROBABILITY
3.4793	2 & 178.	.0319

HETEROGENEOUS VARIANCES (ONE-TAILED TEST)

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	899.6	2	449.81	21.681	.000
ERROR	207.5	10	20.747		
TOTAL	1107.	12			

OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL

GAMES AND HOWELL MODIFICATION OF
TUKEY-KRAMERs HONESTLY SIGNIFICANT DIFFERENCE TEST
(WITH THE STUDENTIZED RANGE STATISTIC)

GROUPS COMPARED	CALCULATED TEST VALUE	DEGREES OF FREEDOM	PROBABILITY	CONCLUSION
5. % ONE-TAILED TEST				
1 AND 2	-5.75	5	.0220	SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 2	5.75	5	.0220	SIGNIFICANT
5. % ONE-TAILED TEST				
1 AND 3	-5.98	2	.0927	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
1 AND 3	5.98	2	.0927	NOT SIGNIFICANT
5. % ONE-TAILED TEST				
2 AND 3	-4.18	2	.1727	NOT SIGNIFICANT
5. % TWO-TAILED TEST				
2 AND 3	4.18	2	.1727	NOT SIGNIFICANT

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Day: 7 / FEMALES

Group-no.: 1

9.000	10.000	6.000	8.000	9.000

MEDIAN= 9.000 MEAN= 8.400 STD = 1.517

Group-no.: 2

11.000	9.000	13.000	10.000	6.000

MEDIAN= 10.000 MEAN= 9.800 STD = 2.588

NOT ENOUGH GROUPS FOR BOXs TEST

CALCULATED F	DEG. OF FREEDOM	PROBABILITY
2.9130	4. & 4	.1628

HOMOGENEITY OF VARIANCES

ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

SOURCE	SS	DF	MS	F	PROB
TREATMENT	4.900	1	4.9000	1.089	.329
ERROR	36.00	8	4.5000		
TOTAL	40.90	9			

NO OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL
NO STATISTICAL DIFFERENCE BETWEEN THE GROUPS

ONE-WAY ANALYSIS OF VARIANCE PROGRAM : ANOVA

Analysis of Day: 14 / FEMALES

Group-no.: 1

14.000	10.000	11.000	15.000	10.000
MEDIAN=	11.000	MEAN=	12.000	STD = 2.345

Group-no.: 2

14.000	8.000	7.000	11.000	.000
MEDIAN=	8.000	MEAN=	8.000	STD = 5.244

NOT ENOUGH GROUPS FOR BOXs TEST

CALCULATED F	DEG. OF FREEDOM	PROBABILITY
5.0000	4. & 4	.0755

HOMOGENEITY OF VARIANCES

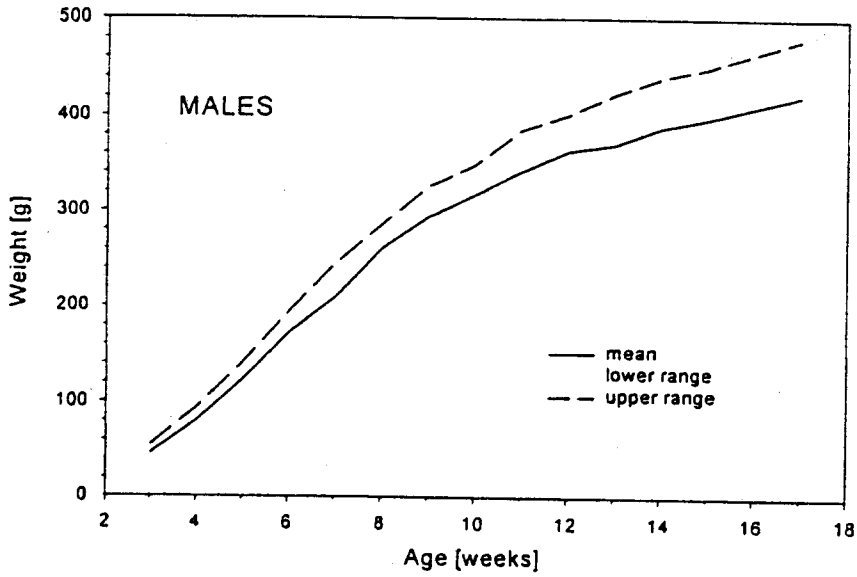
ONE-WAY CLASSIFICATION ANALYSIS OF VARIANCE

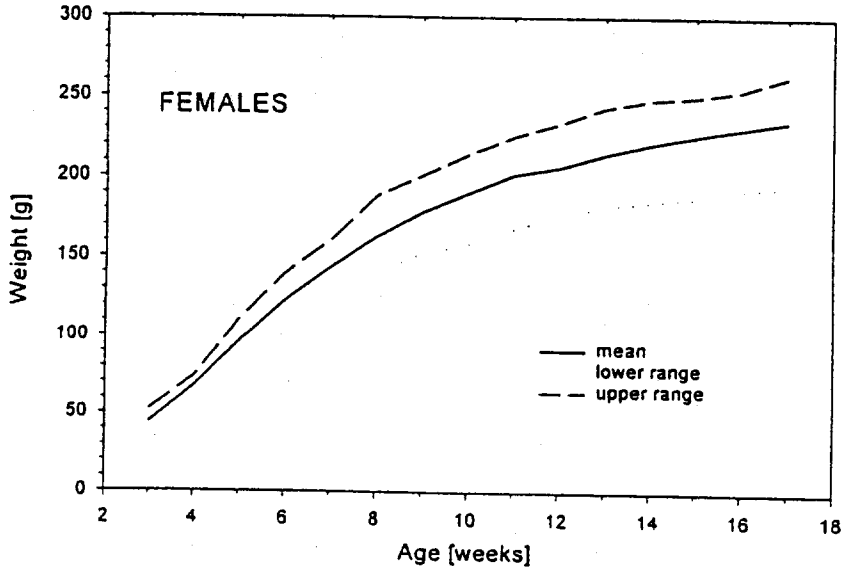
SOURCE	SS	DF	MS	F	PROB
TREATMENT	40.00	1	40.000	2.424	.156
ERROR	132.0	8	16.500		
TOTAL	172.0	9			

NO OVERALL SIGNIFICANCE AT 5.% (ONE-TAILED) LEVEL
NO STATISTICAL DIFFERENCE BETWEEN THE GROUPS

Body Weights / Age-Body Weight Reference Data

(data from Harlan-Winkelmann as of April 1995; n = 30 per sex)





Clinical Observations[1]

[1] Truncated characters: see next set of tables

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Bradypnea

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	5	0	5	0	4	1	5	1	4	0	5	0	2	3	5
1	1	4	0	5	1	4	0	5	1	4	0	5	1	1	1	3
2	3	1	0	5	1	3	0	5	4	0	0	5	1	2	0	3
3	1	0	0	5	3	0	0	5	0	0	0	5	1	2	0	3
4	1	0	0	5	3	0	0	5	0	0	0	5	1	1	0	2
5	1	0	0	5	1	0	0	5	0	0	0	5	0	1	0	2
6	1	0	0	5	1	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Nostrils reddened and encrusted

	Target Concentration - mg/m3 air															
Sex	750 M				2000 M				750 F				2000 F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	3	0	5	0	1	0	5	0	1	0	3
2	0	0	0	5	0	1	0	5	0	0	0	5	0	1	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Dyspnea

	Target Concentration - mg/m3 air															
Sex	750 M				2000 M				750 F				2000 F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Labored breathing pattern

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	3	0	0	5	4	0	0	5	3	0	0	5	5	0	0	5
1	3	1	0	5	2	2	0	5	3	0	0	5	2	1	0	3
2	1	0	0	5	3	0	0	5	0	0	0	5	1	1	1	3
3	0	0	0	5	1	0	0	5	0	0	0	5	1	0	1	3
4	0	0	0	5	1	0	0	5	0	0	0	5	1	0	0	2
5	0	0	0	5	1	0	0	5	0	0	0	5	1	0	0	2
6	0	0	0	5	1	0	0	5	0	0	0	5	1	0	0	1
7	0	0	0	5	1	0	0	5	0	0	0	5	1	0	0	1
8	0	0	0	5	1	0	0	5	0	0	0	5	1	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Motility reduced

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	2	3	0	5	1	3	1	5	1	2	0	5	0	2	3	5
1	2	0	0	5	2	1	2	5	0	0	0	5	0	2	0	3
2	1	0	0	5	2	1	0	5	0	0	0	5	0	1	1	3
3	0	0	0	5	1	0	0	5	0	0	0	5	1	1	0	3
4	0	0	0	5	1	0	0	5	0	0	0	5	1	1	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Atony

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Tremor

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	2	0	0	5	1	4	0	5	1	1	0	5	2	2	1	5
1	0	0	0	5	0	1	2	5	0	0	0	5	0	1	2	3
2	0	0	0	5	1	0	0	5	0	0	0	5	1	0	2	3
3	0	0	0	5	0	0	0	5	0	0	0	5	1	1	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Piloerection

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	1	4	0	5	1	4	0	5	4	1	0	5	0	5	0	5
1	4	1	0	5	0	3	2	5	3	0	0	5	0	2	1	3
2	3	0	0	5	4	1	0	5	2	0	0	5	0	3	0	3
3	3	0	0	5	4	0	0	5	1	0	0	5	1	2	0	3
4	2	0	0	5	2	0	0	5	0	0	0	5	1	1	0	2
5	1	0	0	5	0	0	0	5	0	0	0	5	1	1	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	1	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Salivation

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Ungroomed hair-coat

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	2	1	1	5	0	1	4	5	1	2	0	5	1	0	2	3
2	2	0	0	5	2	3	0	5	3	0	0	5	1	1	1	3
3	2	0	0	5	2	1	0	5	3	0	0	5	2	0	1	3
4	0	0	0	5	0	0	0	5	0	0	0	5	1	0	1	2
5	0	0	0	5	0	0	0	5	0	0	0	5	1	0	1	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Pupils dilated

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	1	5	0	3	2	5	0	2	0	5	1	1	3	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Palpebral narrowing

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	1	2	0	5	0	0	0	5	1	3	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Pallor

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	1	4	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Stilted gait

	Target Concentration - mg/m3 air															
	750				2000				750				2000			
Sex	M				M				F				F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	3	0	5	0	0	0	5	1	0	1	3
2	0	0	0	5	2	0	0	5	0	0	0	5	1	1	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	1	1	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Increased muscle tone

	Target Concentration - mg/m3 air															
Sex	750 M				2000 M				750 F				2000 F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	3	0	5	0	0	0	5	0	3	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	2	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Body weight loss

	Target Concentration - mg/m3 air															
Sex	750 M				2000 M				750 F				2000 F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
2	0	0	0	5	0	1	0	5	0	0	0	5	0	0	0	3
3	0	0	0	5	0	2	0	5	0	0	0	5	0	1	0	3
4	0	0	0	5	0	1	0	5	0	0	0	5	0	1	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Pupils narrowed

	Target Concentration - mg/m3 air															
Sex	750 M				2000 M				750 F				2000 F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	0	1	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Test compound: YRC 2894 480 SC 05776/0096
Study-no: T6067418

Sign: Lying flattened

	Target Concentration - mg/m3 air															
Sex	750 M				2000 M				750 F				2000 F			
Day	l	m	s	N	l	m	s	N	l	m	s	N	l	m	s	N
0	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	5
1	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
2	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
3	0	0	0	5	0	0	0	5	0	0	0	5	0	1	0	3
4	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
5	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	2
6	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
7	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
8	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
9	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
10	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
11	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
12	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
13	0	0	0	5	0	0	0	5	0	0	0	5	0	0	0	1
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Legend: l = slight, m = moderate, s = severe, N = survivors, M = males, F = females

Gross Necropsy

Individual findings / male rats

Group	Animal No.	Time of death	Sacrificed after	Pathology findings
1	1		14 d	**lung**: isolated foci gray-red
	2		14 d	no observable findings
	3		14 d	no observable findings
	4		14 d	no observable findings
	5		14 d	no observable findings
2	21		14 d	no observable findings
	22		14 d	**lung**: foci gray-red
	23		14 d	no observable findings
	24		14 d	no observable findings
	25		14 d	no observable findings
3	11		14 d	**lung**: foci gray
	12		14 d	no observable findings
	13		14 d	no observable findings
	14		14 d	no observable findings
	15		14 d	no observable findings

Individual findings / female rats

Group	Animal No.	Time of death	Sacrificed after	Pathology findings
1	6		14 d	no observable findings
	7		14 d	no observable findings
	8		14 d	no observable findings
	9		14 d	**lung**: foci gray-red
	10		14 d	**lung**: foci gray-red
2	26		14 d	no observable findings
	27		14 d	**lung**: isolated foci dark-red
	28		14 d	no observable findings
	29		14 d	**lung**: foci white
	30		14 d	no observable findings
3	16	24 h		**lung**: slightly collapsed, areas dark-red
	17	4 d		**lung**: slightly collapsed, areas dark-red **liver**: pale **spleen**: pale **kidneys**: pale
	18	6 d		**lung**: slightly collapsed **stomach**: content black mucus, *mucosa*: areas discoloration black **GI-tract**: content black mucus **esophagus**: content full of bedding
	19		14 d	**lung**: slightly collapsed, foci gray-red
	20	1 d		**lung**: slightly collapsed, dark-red

Chow Specification - Nutrients

Altromin Standard Diets 1320 / Totally Pathogene Free TPF®. ALTROMIN 1320 Rat & Mouse Maintenance Diet has now been successfully used for in excess of 30 years and is normally fed to animals aged 8 weeks or over. The diet should be offered ad libitum together with an ample supply of fresh water. Sealed in polyethylene lined sacks, ALTROMIN 1320 can be passed directly into the SPF facility following surface disinfection. ALTROMIN 1324 consists of 10.0 mm pellets.

Specification of Maintenance Diet Rats/Mice

Nutrients (average % content in the diet)	
Crude protein	19.0
Crude fat	4.0
Crude fiber	6.0
Ash	7.0
Moisture	13.5
Nitrogen-free extract	50.5
Metabolizable Energy (calculated)	
Kcal/kg	2850.0
MJ/kg	11.9

Minerals (average % content in the diet)	
Calcium	0.9
Phosphorus	0.7
Magnesium	0.2
Sodium	0.2
Potassium	1.0

Amino Acids (average % content in the diet)	
Lysine	0.90
Methionine	0.30
Cystine	0.30
Phenylalanine	0.80
Tyrosine	0.60
Arginine	1.10
Histidine	0.40
Tryptophane	0.20
Threonine	0.60
Isoleucine	0.80
Leucine	1.30
Valine	0.90

Trace elements (average mg content in 1 kg diet)	
Manganese	75.0
Iron	180.0
Copper	5.0
Zinc	70.0
Iodine	0.9
Fluorine	15.0

Vitamins (additive in 1 kg diet)	Standard-Diet	Standard-Diet fortified
Vitamin A	15000.0 IU	25000.0 IU
Vitamin D_3	600.0 IU	1000.0 IU
Vitamin E	75.0 mg	125.0 mg
Vitamin K_3	3.0 mg	5.0 mg
Vitamin B_1	18.0 mg	30.0 mg
Vitamin B_2	12.0 mg	20.0 mg

Vitamin B_6	9.0 mg	15.0 mg
Vitamin B_{12}	24.0 mcg	40.0 mcg
Nicotinic acid	36.0 mg	60.0 mg
Pantothenic acid	21.0 mg	35.0 mg
Folic acid	2.0 mg	3.0 mg
Biotin	60.0 mcg	100.0 mcg
Choline	600.0 mg	1000.0 mg
Vitamin C	36.0 mg	60.0 mg

Chow Specification - Impurities

Impurity	Max. acceptable value	LUFA - Limit of detection	Altromin *
Aflatoxine B1 / B2	0.01	0.0025	nng
Aflatoxine G1 / G2	0.01	0.0025	nng
Antibiotic activity	± 0		nng
Arsenic	2.0	0.2	0.3
Fluoride	150.0	5.0	22.0
Mercury	0.1	0.01	0.08
Lead	5.0	0.1	0.37
Cadmium		0.01	0.10
Selenium		0.10	1.0
Tecnazene		0.001	< 0.001
Quintozene		0.001	< 0.001
HCB (Hexachlorbenzene)		0.001	< 0.001
α -HCH		0.001	< 0.001
β -HCH		0.002	< 0.002
τ -HCH	0.1	0.001	0.002
Heptachlor	0.03	0.005	< 0.005
Heptachlorepoxid	0.03	0.005	<0.005
α - Chlordan	0.05	0.005	<0.005
τ - Chlordan	0.05	0.005	<0.005
Aldrin	0.02	0.005	<0.005
Dieldrin	0.02	0.005	<0.005
Endrin	0.02	0.01	< 0.01
o,p - DDE	0.05	0.005	<0.005
p,p - DDE	0.05	0.005	<0.005
o,p - DDD	0.05	0.005	<0.005
o,p - DDT	0.05	0.005	<0.005
p,p - DDD	0.05	0.01	< 0.01
p,p - DDT	0.05	0.01	< 0.01
Methoxychlor		0.01	< 0.01
PCB qual.			nng
Chlorthion		0.01	< 0.01
Disulfothion		0.005	<0.005
Malathion		0.01	< 0.01
Methylparathion		0.005	<0.005
Ethylparathion		0.01	< 0.01
Sulfotepp		0.002	<0.002
Fenthion		0.005	<0.005
Diazinon		0.01	< 0.01
Dibrom		0.02	< 0.02
Dimethoate		0.005	<0.005
Trichlorphon		0.01	< 0.01
Fenitrothion		0.01	< 0.01

* In this study Altromin 1324 was used. 4 is the degree of pelletation.Dimension: ppm

Tap Water Specification

No.	Substance	Limit mg/l	computed as	equivalent mmol/m³	acceptable error of value (± mg/l)
1	Arsenic	0.04	As	0.5	0.015
2	Lead	0.04	Pb	0.2	0.02
3	Cadmium	0.005	Cd	0.04	0.002
4	Chrome	0.05	Cr	1	0.01
5	Cyanide	0.05	CN-	2	0.01
6	Fluoride	1.5	F-	79	0.2
7	Nickel	0.05	Ni	0.9	0.01
8	Nitrate	50	NO_3^-	806	2
9	Nitrite	0.1	NO_2^-	2.2	0.02
10	Mercury	0.001	Hg	0.005	0.0005
11	Polycyclic aromatic carbohyrates - Fluoranthene - Benzo-b-fluoranthene - Benzo-k-fluoranthene - Benzo-a-pyrene - Benzo-(ghi)-perylene - Indeno-(1,2,3-cd)-pyrene	sum 0.0002	C	0.02	0.00004
12	Organochlorine compounds - 1,1,1-Trichlorethane - Trichlorethylene - Tetrachlorethylene - Dichlormethane	sum 0.01	-	-	0.004
	- Tetrachlormethane	0.003	CCl4	0.02	0.001
13	a. Pesticides	indiv-dual com-pound 0.0001	-	-	0.00005
	b. Polychlorinated Polybromated biphenyles and terphenyles	sum 0.0005	-	-	0.0002
14	Antimony	0.01	Sb	0.08	0.002
15	Selenium	0.01	Se	0.13	0.002

Specification of Test Compound

Bayer AG
PF-PM/PPA

03.06.98

Approval of Preparation Sample

Preparation Sample TOX 4627

Sample: *YRC 2894 00480 SC 05776/0096*

Development-No.: *0215336*

Indication: *Insecticide*

Active Ingredients: *1. YRC 2894*

Formulation No.: *0100 based on Formulation No.: 05776/0096*

Origin of sample: *PF-E/FT*

Responsible Analyst: *Dr. Teller* **Laboratory:** *FT-EA*

Analytical Methods: *HPLC, ext. Std.*

Approvals:

TOX	Purity		Approved until	Date of Analysis	Comment
4627-00	*1. 490.6*	*g/l*	*05.11.98*	*05.05.98*	

(H. Baum, PF-PM/PPA)

A reserve sample will be retained.

Reasons for the Revision

Page 8, 30, 34
Aerosol concentration has been re-calculated based on gravimetric analysis but considering only non-volatile constituents of YRC 2894 (989 mg/m^3 and 2199 g/m^3 = actual concentrations).
Footnote: ...gravimetrically determined concentrations... changed in ...actual concentrations ...

Page 28, 40
Actual concentrations of the test-substance were added to the gravimetrically determined concentrations.

Page 16
Dispersion pressure approximately 150 kPa (instead of 600 kPa)

Page 24
Controls were exposed to aerosolized water (instead of conditioned air)

Editorial changes
Page 12, 16, 18, 28, 29, 30, 34, 56-63
Addition of developmental number. „Purity" interchanged with „Content"
Levatit water
Control rats were exposed in a chamber consisting of 2 segments. All technical settings were adjusted accordingly (Sentence deleted)
Actualization of legend - deletion of NMAD
The lower gravimetrical concentration compared with the nominal concentrations are attributed ~~to the incomplete aerosolization of water particles and/or~~ to the evaporation of water.
Onset ~~and Duration~~ of Mortality
As determined by gravimetric evaluations the scheduled test-substance concentrations were nearly met for the low-dose groups. The scheduled test substance concentration in high-dose animals was not met but it was in the range of the maximum technically attainable concentration (Sentence deleted).
In the light of the results of the analytical concentrations of active ingredients in the exposure atmosphere and the clinical findings toxicity appears to be related to the test substance (Sentence deleted).
„Kontrolle" translated into „Control" and mg/cbm in mg/m^3

End of Report