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SOLVAY FLUORIDES

A SUBSIDIARY OF SOLVAY CHEMICALS, INC.

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March 4, 2004

via Overnight Courier

Attn: Document Control Officer 7407
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8EHQ-04-15528

CONTAINS NO CBI

Re: Solvay Fluorides---TSCA Section 8(e)---Sodium Hexafluoroaluminate (CAS No. 13775-53-6---90 Day Repeat Dose Inhalation Study in Rats (snout only exposure))

Dear Sir/Madam:

This letter is being submitted by Solvay Fluorides, LLC ("Solvay Fluorides") pursuant to Section 8(e) of the Toxic Substances Control Act (TSCA). Enclosed is the 90 Day Repeat Dose Inhalation Study in Rats.

This study was performed at Huntingdon Life Sciences, Huntingdon Research Centre, Huntingdon, England, to assess the response of rats to inhalation exposure, snout only, to a particulate aerosol generated from Cryolite (sodium hexafluoroaluminate). Groups of rats were exposed for 6 hours a day, 5 days a week for 13 consecutive weeks to study mean analyzed concentrations of 0.21, 1.04 and 4.6 mg/m^3 Cryolite in air.

Pulmonary inflammatory lesions associated with exposure to Cryolite were seen in a majority of animals receiving Cryolite at the High dose and, to a lesser degree, in some animals from the Intermediate dose group.

No treatment-related changes were seen in animals from the Low dose Cryolite group.

The treatment related changes observed in animals in the High and Intermediate dose groups have, with the exception of the presence of small foci of brown pigmented alveolar macrophages, resolved after the conclusion of the recovery/withdrawal period of 13 weeks.

2004 MAR 15 AM 11:20

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Solvay Fluorides, LLC
A Subsidiary of Solvay Chemicals, Inc.
3333 Richmond Avenue, Houston, Texas 77098
Tel: 713.525.6700 FAX: 713.525.7805
www.solvaychemicals.us

273682

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Brown pigment containing macrophages were seen in the tracheobronchial and mediastinal lymph nodes of a proportion of High dose Cryolite rats.

Animals from Group 2 (NaF) comparative control group exhibited laryngeal epithelial hyperplastic and inflammatory changes. Laryngeal changes were not seen in animals receiving Cryolite.

The Low dose group (0.21 mg/m^3) was considered to be the no-effect level for exposure to Cryolite in this study.

If there are any questions please feel free to contact me at 713-525-6570.

Sincerely,

Vance Erickson
Sr. Vice President
Solvay Fluorides, LLC

Enclosure
rdw04002

Solvay Fluorides, LLC
A Subsidiary of Solvay Chemicals, Inc.
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SODIUM HEXAFLUOROALUMINATE (Natriumaluminiumfluorid)
CAS No. 13775-53-6

B G No. 107

90 DAY REPEAT DOSE INHALATION STUDY IN RATS
(snout only exposure)

Sponsor

BG Chemie,
Kurfürsten-Anlage 62,
69115 Heidelberg,
GERMANY.

Sponsor's representative

Dr. G Martens

Research Laboratory

Huntingdon Life Sciences Ltd.,
P.O. Box 2,
Huntingdon,
Cambridgeshire,
PE18 6ES,
ENGLAND.

Report issued: 26th November 1997

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APPENDICES (continued)

COMPLIANCE WITH GOOD LABORATORY PRACTICE STANDARDS

The study described in this report was conducted in compliance with the following Good Laboratory Practice standards and I consider the data generated to be valid.

Good Laboratory Practice, The United Kingdom Compliance Programme, Department of Health 1989, and subsequently the United Kingdom Good Laboratory Practice Regulations 1997, Statutory Instrument No. 654.

EC Council Directive, 87/18 EEC of 18 December 1986, (No. L 15/29).

Good Laboratory Practice in the testing of Chemicals OECD, ISBN 92-64-12367-9, Paris 1982, subsequently republished OECD Environment Monograph No. 45, 1992.

United States Environmental Protection Agency, (TSCA), Title 40 Code of Federal Regulations Part 792, Federal Register, 29 November 1983 and subsequent amendment Federal Register 17 August 1989.

United States Environmental Protection Agency (FIFRA), Title 40, Code of Federal Regulations Part 160, Federal Register, 29 November 1983 and subsequent Amendment, Federal Register 17 August 1989.

Japan Ministry of International Trade and Industry, Directive 31 March 1984 (Kanpogyo No. 39 Environmental Agency, Kikyoku No. 85 MITI).

26 November 1997

Derek W. Coombs, B.Sc.,
Study Director,
Huntingdon Life Sciences Ltd.

Date

QUALITY ASSURANCE STATEMENT

This report has been audited by Huntingdon Life Sciences Quality Assurance Department (Huntingdon). The methods, practices and procedures reported herein are an accurate description of those employed at Huntingdon during the course of the study. Observations and results presented in this final report form a true and accurate representation of the raw data generated during the conduct of the study at Huntingdon.

Inspections were made by the Quality Assurance Department of various phases of the study as conducted at Huntingdon and described in this report. The dates on which the inspections were made and the dates on which findings were reported to the Study Director and to Management, Huntingdon Life Sciences are given below.

Phase of Study	Date of Inspection	Date of Reporting
Protocol Review	-	24 July 1996
Pre-experimental Period	6 August 1996	6 August 1996
Experimental Period	15 August 1996	19 August 1996
	3 October 1996	3 October 1996
	4, 5 & 7 November 1996	7 November 1996
	9 January 1997	9 January 1997
	6 February 1997	6 February 1997

Date of reporting audit findings to the Study Director and Management — 11 November 1997

Mark Somerset,
Audit Team Supervisor,
Department of Quality Assurance,
Huntingdon Life Sciences Ltd.

25 November 1997
Date

RESPONSIBLE PERSONNEL

STUDY MANAGEMENT

Derek W. Coombs, B.Sc.,
Study Director.

Mario Bannerman, H.N.D.,
Head of Inhalation Toxicology.

TOXICOLOGY

Colin J. Hardy, B.Sc., Ph.D., M.I.Biol., C.Biol.,
Dip.R.C.Path. (Toxicology),
Principal Toxicologist.

CLINICAL PATHOLOGY

David Crook, B.Sc., Ph.D.,
Head, Department of Clinical Pathology.

OPTHALMOSCOPIC EXAMINATION

David P Buist., B.Vet.Med., M.R.C.V.S.,
Veterinary Director and Principal Veterinarian,
Department of Veterinary Services.

PATHOLOGY

Chirukandath Gopinath, B.V.Sc., M.V.Sc., Ph.D., F.R.C.Path.,
Director of Pathology.

SUMMARY

This study was performed at Huntingdon Life Sciences, Huntingdon Research Centre, Huntingdon, England to assess the response of rats to inhalation exposure, snout only, to a particulate aerosol generated from Cryolite. Groups of rats were exposed for 6 hours a day, 5 days a week for 13 consecutive weeks to study mean analysed concentrations of 0.21, 1.04 and 4.6 mg/m^3 Cryolite in air.

An additional group of rats were exposed to a study mean analysed concentration of 5.7 mg/m^3 Sodium Fluoride (NaF), acting as a comparative control. A fifth group of rats was exposed to air only.

No effects of treatment were evident in clinical signs, bodyweight gain, food or water consumption. Haematology, biochemistry and urinalysis investigations did not reveal any treatment-related differences.

The following were considered treatment-related:

Urinary fluoride

Increased for Groups 2 (NaF) and 5 (High dose Cryolite).

Organ weights

Increased lung weights were present in Group 5 (High dose Cryolite) at termination. A slight increase above Air Control values remained at the end of the withdrawal period.

Microscopic pathology

Pulmonary inflammatory lesions associated with exposure to Cryolite were seen in a majority of animals receiving Cryolite at the High dose and, to a lesser degree, in some animals from the Intermediate dose group.

No treatment-related changes were seen in animals from the Low dose Cryolite group.

The treatment related changes had, with the exception of the presence of small foci of brown pigmented alveolar macrophages, resolved after the conclusion of the recovery/withdrawal period. Brown pigment containing macrophages were seen in the tracheobronchial and mediastinal lymph nodes of a proportion of High dose Cryolite rats.

Animals from Group 2 (NaF) comparative control group exhibited laryngeal epithelial hyperplastic and inflammatory changes. Laryngeal changes were not seen in animals receiving Cryolite

The Low dose group (0.21 mg/m^3) was considered to be the no-effect level for exposure to Cryolite in this study.

INTRODUCTION

The purpose of this study, performed at Huntingdon Life Sciences Ltd, Huntingdon Research Centre, England, was to assess the response in rats to repeat inhalation exposures, by snout-only administration, of the test substance, 5 days a week for 13 consecutive weeks. Additional rats were included in the Air control, NaF comparative control and High dose Cryolite groups and were held for a 13 week withdrawal period following the last exposure.

Satellite animals (male only) were included for determination of chromosomal aberrations at the end of 13 weeks exposure. (Six male rats were held but not exposed during the 13 weeks of exposures, at the end of which time they were dosed intraperitoneally with Cyclophosphamide as a positive control).

The fluoride and aluminium levels in samples of urine, bone (femur) and teeth (lower incisors) were determined in main and withdrawal group rats. Samples were pooled from each cage of rats.

The study was designed with reference to national and international guidelines to meet the requirements of regulatory authorities for the toxicity testing of industrial chemicals. In particular the study complied with OECD Guidelines for the testing of chemicals No. 413.

The test substance was administered by inhalation, a possible route for accidental exposure in man. The rat is the species of choice due to regulatory requirements and the strain was selected on account of the availability of comprehensive background data, relating to clinical and pathological parameters, at our laboratories.

Dose levels were selected, in consultation with the sponsor, on the basis of data from two 2 week inhalation studies BGH 47/932236 and BGH 56/942492.

All specimens, raw data and other documents generated during the course of this study and a copy of this final report have been lodged in the Huntingdon Life Sciences Archives, Huntingdon, Cambridgeshire. Material will not be discarded or released from these laboratories without the prior consent of the Sponsor.

Notes: Satellite rats were included for determination of chromosomal aberrations. Although clinical signs and bodyweight were recorded for those animals, they were not included in the toxicological evaluation. The data are retained in the study file but have not been reported.

RELEVANT STUDY DATES

Approved by:	
Study Director	19 June 1996
Management	19 June 1996
Study Sponsor	8 July 1996
Animals arrived at Huntingdon Life Sciences:	24 July 1996
Exposures commenced:	8 August 1996
Haematology/biochemistry:	
Week 13	5 November 1996
Urine collection:	
Week 13	5 November 1996
Week 26	5 February 1997
Terminal kill:	7/8 November 1996
Withdrawal kill:	6/7 February 1997

TEST SUBSTANCE

	Test substance	Comparative control substance
Chemical name:	Sodium hexafluoroaluminate (Natriumaluminiumfluorid)	Sodium fluoride (Natriumfluorid)
Common name:	Cryolite	-
Referred to as in this report:	Cryolite	NaF
CAS number:	13775-53-6	7681-49-4
BG Chemie No:	107	-
Presentation:	Powder	Powder
Storage conditions:	Dry at room temperature in the dark	Dry at room temperature in the dark
Batch/Product no.:	Partie no. 2	00.00002151
Purity:	98.9%	100.8%
Expiry date:	10 September 1997	29 November 1999
Date received:	17 December 1992	21 February 1996
Supplier:	Bayer AG Zentrale Forschung und Entwicklung Zentrale Analytik 5090 Leverkusen Bayerwerk Germany	Bayer AG ZF - Forschungdienste Building Q18 D-51368 Leverkusen Germany

EXPERIMENTAL PROCEDURE

ANIMALS

Two hundred and nine (117 male and 92 female) rats aged approximately 6 weeks, of the Crl: CD® BR Sprague-Dawley strain were obtained from Charles River (UK) Limited, Manston Road Margate, Kent, England, on 24 July 1996.

All of the rats were examined upon receipt and appeared to be in good health.

Five male and five female rats (185 - 189 male and 190 - 194 female), designated as a health check were selected at random and sacrificed. At necropsy a detailed macroscopic examination was performed. No abnormalities were present and no evidence of infectious disease was seen.

Pre-dose ophthalmic examination took place 25 July 1996, prior to allocation. Four male rats were rejected due to eye lesions and replaced. This reduced the number of male rats required in the reserve group.

Allocation took place on 25 July 1996. Individual bodyweights were processed using a computer program which selected 160 rats (80 male and 80 female) for allocation to 5 groups such that the group mean bodyweights were approximately equalised. In addition, 24 male rats were selected for allocation to 4 satellite groups used in the determination of chromosomal aberrations, including a positive control dosed with intraperitoneal cyclophosphamide.

All animals were then uniquely identified by numbers tattooed into the ear pinnae. For numbers exceeding 99, the number 1 was tattooed on the right hind foot of the rat. Surplus rats (2 female) were killed on the day of allocation.

The identification of individual rats in all groups, together with the initial group mean weights at allocation were as follows:

Group	Rat numbers					Group mean Body weights (g)	
	Main study		Withdrawal		Satellite		
	Male	Female	Male	Female	Male		
1 (Air control)	1 - 10	81 - 90	11 - 20	91 - 100	161 - 166	142	143
2 (NaF)	21 - 30	101 - 110	31 - 40	111 - 120	167 - 172	143	143
3 (Low dose Cryolite)	41 - 50	121 - 130				142	143
4 (Inter dose Cryolite)	51 - 60	131 - 140				142	143
5 (High dose Cryolite)	61 - 70	141 - 150	71 - 80	151 - 160	173 - 178	142	142
6 (ip cyclophophamide)					179 -184	146	
Reserve	A - D	F - J					

Of the remaining animals, 4 male and 5 female were assigned to the reserve group. These were retained as potential replacements during the acclimatisation period. At pre-exposure ophthalmoscopy 4 male rats were rejected with adverse eye lesions. These rats were replaced by reserve males. Following the commencement of exposures remaining reserve rats (all female) were killed.

ACCOMMODATION

The rats were housed 5 (Main study and Withdrawal) or 6 (Satellite) of the same sex to a cage in suspended stainless steel cages fitted with mesh front, back and floor with stainless steel sheet sides. Each cage was 35 cm wide, 53 cm long and 25 cm high. Plastic trays lined with absorbent paper, were placed below each cage to collect animal excreta. The paper was changed daily. Clean cages were introduced at intervals throughout the study. During the period of exposures the rats were kept in a single room (Building Y14, room 008) and, additionally, after the start of the exposure period, each group was positioned on an individual cage battery. Each battery was housed in a separate ventilated cabinet within the holding room in order to avoid the possibility of inhalation of test material from the fur of rats in other groups. Exposure took place in the same room. The temperature and relative humidity of the holding room were recorded using a Kent Clearspan chart recorder. The study holding room temperature and relative humidity were set to be maintained within limits of 21 ± 3°C and 55 ± 15% respectively. Recorded ranges are shown in Appendix 2.

Lighting was controlled to give 12 hours light (0730 - 1930 hours) and 12 hours dark per 24 hours.

DIET

While in their cages, all rats had access to a weighed quantity of standard quality-controlled laboratory rat food (SDS Rat and Mouse No. 1 SQC modified maintenance diet, Special Diets Services, Witham, Essex).

There was no information available to indicate that any non-nutrient substance likely to influence the effect of the test compound could reasonably be expected to be present in the diet. Analyses were made on all batches of diet used to establish levels of basic nutrients and of specific substances and micro-organism likely to have been present in the feed components and which, if in excess of specified amounts, might have an undesirable effect on the test system (see Appendix 1).

The analytical data have been lodged in Huntingdon Life Sciences Archives.

Tap water was available from moulded polypropylene water bottles at all times while the rats were in the cages. The water bottles were rinsed and refilled daily and thoroughly cleaned at intervals during the study.

There was no information available to indicate that any substance likely to influence the effect of the test system could reasonably be expected to be present in the drinking water.

Results of the routine physical and chemical analyses of water at source (sampling point, Grafham Final Water) as conducted by the supplier, Anglian Water Services Ltd, have been made available to Huntingdon Life Sciences. Additionally, levels of specified substances known to be present from time to time in local water and which, if in excess of the maxima recommended for (human) drinking water, might have an undesirable effect on the test system, are determined in the tap water at intervals. A list of the principal determinands is given in Appendix 1.

The analytical data have been lodged in Huntingdon Life Sciences Archives.

Approximately 20 g of each batch of diet used during the study, together with approximately 20 ml water, were collected at 2 weekly intervals and stored in a refrigerator for possible future analysis of Fluoride and Aluminium.

ADMINISTRATION

A particulate aerosol generated from Cryolite or sodium fluoride was administered to rats by snout only exposure, 6 hours a day, 5 days a week for 13 consecutive weeks. Control rats received air only.

Withdrawal rats in Groups 1 (Air control), 2 (NaF) and 5 (High dose Cryolite) were maintained in their holding cages for a 13 week period following the last exposure. For logistic reasons these rats were transferred to Room 010 building Y11 for the period of withdrawal.

Due to technical problems exposures 63 and 65 were 5 hours duration. This was considered not to have effected the scientific integrity of the study.

INHALATION EXPOSURE SYSTEM AND PROCEDURE

EXPOSURE SYSTEM

The exposure system employed consisted of a dust generator to produce an aerosol from the powder supplied, an ADG snout-only inhalation chamber and rat restraining tubes. Accessories included air supply and extract lines which attached to the top and bottom of the chamber respectively and an air supply to the dust generator. A filtration system was incorporated in the extract line.

A line drawing of an exposure system is shown in Figure 1. The component parts of the system are described in further detail below:

Wright dust feed

The test atmosphere was produced using a Wright dust feed mechanism (WDF). In this device a canister packed with the test substance is rotated against a scraper blade. Dried, compressed air passed through the dust feed and carried the powder removed from the dust cake out into the air supply and into the exposure chamber. A line drawing of the WDF mechanism is shown in Figure 2.

Different concentrations of the powder are produced by selection of different gearing ratios available for use with this device.

Inhalation chamber

ADG snout-only inhalation chamber (ADG Developments Ltd, Hitchin, Hertfordshire, England). A modular apparatus of aluminium alloy construction comprising a base unit, a variable number of animal exposure sections each having 20 exposure ports and a top section incorporating a central aerosol inlet surrounded by a tangential air inlet.

Each chamber used on the present study was assembled using 3 animal exposure sections, identified as levels 1 (top) to 3 (bottom), and formed a 28 cm diameter cylinder with a volume of approximately 47 litres. During dosing, each chamber was housed in an enclosed ventilated cabinet to avoid cross contamination.

Rat restraining tubes

Moulded polycarbonate tubes tapered at one end to allow the snout only to project from the tapered end. The other end was closed by insertion of an expanded plastic bung. A push-rod passed through the centre of the bung and was adjustable to maintain the position of a rat during restraint. Tubes were attached to a chamber by means of push-fit "O" ring seals located in the exposure ports of the animal exposure sections.

Air supply and extract

Each exposure system was operated with an air extract attached to the base of the inhalation chamber. A supply of clean, dried compressed air was used to operate the WDF. A supplementary air supply to the tangential air inlet at the top of the chamber was used to balance the chamber airflows. An air extract line normally includes a silica gel column to remove excess moisture from the air and a filtration system to remove particulate matter.

Air supplies were provided by a compressor. The air was filtered to remove any residual particulate and was dried (dew point ~2°C). Air extract was provided by vacuum pumps. Air supplies and extracts were monitored using in-line tapered tube rotameters.

The in-line flowmeters were calibrated against high quality tapered tube rotameters measuring the free flow of air at the points of attachment of the supply and extract lines to each chamber, and from the outlet of each WDF. Details of the system operating parameters are presented in Appendix 3.

Five exposure systems were used, one for each group.

The procedure followed for each group was similar except that, for the control group, an empty WDF canister was attached to the WDF. As such, the rats in the control group received air only.

Therefore, in the following description, the comments relating to the use of packed containers with the WDF applied to the test groups only.

PROCEDURE

The rats were removed from their cages and placed into restraining tubes which were colour coded for the treatment group and numbered for the cage from which an animal was drawn. A separate chamber was used for each group. For each group, all the rats were loaded on all 3 levels of the appropriate chamber. Unused animal exposure ports were sealed with blanking plugs. The air supply and extract lines were connected to the chamber and WDF.

With the animals in position on the chamber, the container of the WDF was advanced manually until a trace of dust was seen to emerge from the outlet of the WDF. The gearing of the WDF was then engaged and the instrument switched on. Exposure commenced from the moment when the WDF was switched on. Operation of the exposure system was monitored frequently during the period of exposure.

Samples for the determination of total chamber concentration and particle size distribution were taken during each six- hour exposure. Records of chamber temperature were made together with the reaction, if any, of exposed rats.

On completion of the 6-hour exposure period, the WDF in use was switched off, the air supplies disconnected and one of the blanking plugs removed from the chamber. The system was then allowed to clear for approximately 5 minutes before the rats were removed from the restraining tubes and returned to their holding cages. The restraining tubes were then washed.

The nominal rate of aerosol production was confirmed by weighing the WDF containers and contents before and after use. The WDF containers were removed at appropriate intervals during the study (Table 3).

TARGET CONCENTRATIONS

The target concentrations were:

Group	Designation	Concentration (mg/m^3)
2	NaF	6.0
3	Low dose Cryolite	0.2
4	Inter dose Cryolite	1.0
5	High dose Cryolite	5.0

The target concentrations in this study were selected following review of the results obtained from two 2 Week studies performed at Huntingdon Life Sciences (Study number BGH 47/932236 and BGH 56/942492) and in consultation with the Sponsor.

EXPOSURE CHAMBER CONDITIONS

Chamber concentration of Cryolite and sodium fluoride

During the exposure the chamber atmosphere was sampled for total particulate concentration on two (Group 3) or three occasions (Groups 2, 4 and 5). Samples were taken approximately 1 and 3 hours into the exposure for Group 3 and 1, 3 and 4.5 hours into the exposure for Groups 2, 4 and 5. The reduced frequency of sampling was a direct result of the low concentration at the low dose exposure level and the necessity to collect enough particulate for accurate weighing. Gravimetric determination only was possible in this investigation.

Samples for total particulate were collected using Whatman GF/A glass fibre filters and air was drawn through each filter in its holder using a vacuum pump. Volumes of each sample removed were measured by an in-line Wet-type gas meter.

Particle size distribution

Samples of chamber atmosphere were removed from each exposure chamber on one occasion each week for determination of particle size distribution of Cryolite.

It was intended to use a Cyclone sampler (Cassella Ltd., Kempston, Bedford, England) in order to determine the particle size distribution of the aerosol. This 2-stage device would have enabled daily measurement defining respirability as the proportion of particulate less than 7 μm, and would have enabled relatively high frequency sampling at the extremely low particulate levels used on the study.

However, due to technical problems during the first exposure, associated with the weighing procedure of the appropriate components of the Cyclone, it became obvious that use of this device was not a practical proposition. Subsequent samples were taken using a Marple Model 296 Personal Cascade impactor sampler.

As a result no particle size data was obtained for Group 3 (Low dose Cryolite) during Week 1 of dosing, as the required time necessary using the Marple in order to collect sufficient particulate on each stage for accurate weighing was 5 days. This lack of data was considered not to affect the study integrity. Using the Marple device samples removed for particle size determinations were limited to once per week for each group. However, using this device enabled calculation of the Mass Median Aerodynamic Diameter (MMAD).

The Marple Model 296 Personal Cascade impactor was used at a flow rate of 2 litres per minute and the collection characteristics at this flow rate were as follows:

Impactor stage	Cut point [a] (μm)
3	9.8
4	6.0
5	3.5
6	1.55
7	0.93
8	0.52
Filter	0

[a] Aerodynamic equivalent particle diameter for spherical particles of unit mass density in air at 25°C and 1 atmosphere.'

The Cryolite deposited on each stage was calculated by weight difference. The mass median aerodynamic diameter of the particulate was calculated by linear regression analysis of the probit of the cumulative percentage of the total particles collected, smaller than the cut-point of each stage, against the logarithm of the cut-point of each stage.

Nominal concentration

The nominal concentration of Cryolite or NaF in each chamber was calculated from the total weight of powder lost from each dust container at each level of exposure, during the period in which each canister was in use, divided by the total airflow through the system over the same period.

$$\frac{\text{Amount of test substance used in 6 hours (mg)}}{\text{Total chamber airflow in 6 hours } (m^3)} = mg / m^3 \text{ in air}$$

Chamber air flow

The air flow into each chamber was monitored continuously using tapered tube flow meters and recorded at approximately 30-minute intervals throughout each exposure.

Chamber temperature

The temperatures of each chamber were recorded at 30-minute intervals throughout each exposure.

CLINICAL SIGNS

During exposure

Clinical signs during exposure are normally recorded as a group response where all visible animals appeared to be responding similarly or a proportion were affected. However, the range of clinical signs was severely limited by the method of restraint used and were limited to any accidental tube restraint deaths.

At other times

All rats were individually examined at least once each week and an entry made on individual clinical signs sheets even if abnormal behaviour or reactions were not seen. In addition, at least twice each day, (at loading and unloading on exposure days), the animals were examined for any abnormal signs. Any abnormal signs were recorded on the individual sheets.

BODYWEIGHT

Each rat was weighed for allocation to groups, then weekly commencing 1 week before the start of exposures and continuing throughout the study.

In addition, the weight of each rat at necropsy was recorded.

FOOD CONSUMPTION

The quantity of food consumed by each cage of main study or withdrawal rats was recorded weekly commencing 1 week prior to the start of exposures until the end of the study.

The amount of food consumed was determined by weight loss from the food hoppers.

WATER CONSUMPTION

The quantity of water consumed by each cage of main study or withdrawal rats was recorded daily, commencing 1 week prior to the start of exposures until the end of the study.

The amount of water consumed by each cage of rats was determined daily by weight loss from the water bottles.

OPHTHALMIC EXAMINATION

The eyes of all rats submitted for allocation were examined once before the start of exposure using a Keeler indirect ophthalmoscope. Similarly, all rats in Groups 1, 2 and 5 were examined during Week 13 (prior to exposure on the day).

Prior to examination, the pupils of each animal were dilated using a Tropicamide ophthalmic solution (0.5% Mydriacyl).

LABORATORY INVESTIGATIONS

Sample collection

Blood samples were collected from all main study animals on one occasion during Week 13. Urine samples were collected from all main study animals on one occasion during Week 13 and from surviving withdrawal animals during Week 26 (Week 13 of withdrawal).

Overnight urine samples were collected from rats placed in individual urine cages between approximately 1700/1730 and 0800/0830 hours the following day. Food and water were withheld from rats during urine collection. Samples collected during the withdrawal phase of the study (Week 26) were only used for analysis of aluminium and fluoride.

Prior to the collection of overnight urine samples, the individual metabolism cages were washed with a solution of nitric acid in deionised water (1:20 v/v) to remove any fluoride or aluminium contamination that may have been present. This was followed by rinsing with deionised water followed by air drying.

The glass urine separators used for collection were also rinsed in nitric acid solution and rinsed with deionised water. The separators and vials were heat dried in an oven.

Samples of venous blood were obtained from the orbital sinus using sterile glass pipettes while the rats were maintained under light ether anaesthesia. Water was returned to the cages for approximately 1 hour before removal of blood samples.

The blood samples collected were divided into tubes containing the following anticoagulants:

EDTA - for haematological investigations
Heparin - for biochemical investigations

Following removal of blood samples all the rats were allowed access to food and water in their holding cages prior to exposure on the day.

The haematological, blood biochemical and urinalysis investigations performed are listed below, together with an abbreviated title (for use in appendices and tables), methods and the units of measurement.

Haematology

The following estimations were performed using a Bayer-Technicon H*1E haematology analyser:

	Units
Packed cell volume (PCV)	%
Haemoglobin (Hb)	g/dl
Red cell count (RBC)	$\times 10^{12}$/l
Absolute indices calculated as follows:	
Mean corpuscular haemoglobin concentration (MCHC) Hb (g/dl) × 100 ÷ PCV (%)	g/dl
Mean corpuscular volume (MCV) PCV (%) × 10 ÷ RBC ($\times 10^{12}$/l)	fl
Mean corpuscular haemoglobin (MCH) Hb (g/dl) × 10 ÷ RBC ($\times 10^{12}$/l)	pg
Total white cell count (WBC Total)	$\times 10^{9}$/l
Differential WBC count (Diff)	
Neutrophils (N) Lymphocytes (L) Eosinophils (E) Basophils (B) Monocytes (M) Large unstained cells (LUC)	$\times 10^{9}$/l

Cell morphology: the most common morphological changes (anisocytosis, micro/macrocytosis, variation in colour, hypo/hyperchromasia, left shift, atypical/blast cells) were recorded as follows:

-	=	no abnormalities detected
+	=	slight
++	=	moderate
+++	=	marked

In the case of atypical/blast cells, or other abnormalities, confirmation or a written description from a blood film was made.

Platelet count (Plts)	$\times 10^{9}$/l
Reticulocyte count (Retic) - Method of Dacie, J.V. and Lewis, S.M. (Practical Haematology, 1966, 3rd edition p.28)	% (of red cells)

Biochemistry

The following parameter was analysed with a Roche Cobas centrifugal analyser, using the appropriate BCL test kit:

	Units
Creatine phosphokinase (CPK), also known as `creatine kinase' - reaction temperature 30°C	mU/ml

The following parameters were analysed with a Hitachi 737 clinical chemistry analyser, using standard Hitachi 737 methodology:

Total protein	g/dl
Albumin (Alb)	g/dl
Globulin (Glob) - By subtraction Total Protein (g/dl) minus Albumin (g/dl)	g/dl
Urea Nitrogen (Urea Nitr)	mg/dl
Alkaline phosphatase (AP) Reaction temperature 30°C	mU/ml
Total bilirubin	mg/dl
Creatinine	mg/dl
Sodium (Na)	mEq/l
Potassium (K)	mEq/l
Calcium (Ca)	mEq/l
Inorganic phosphorus (P)	mEq/l
Chloride (Cl)	mEq/l
Cholesterol (Chol)	mg/dl
Glucose - hexokinase mediated	mg/dl
Glutamic-pyruvic transaminase (GPT), also known as `alanine aminotransferase' Reaction temperature 30°C	mU/ml
Glutamic-oxaloacetic transaminase (GOT), also known as `aspartate aminotransferase' Reaction temperature 30°C	mU/ml
Gamma-Glutamyltransferase (γGT) Reaction temperature 30°C	mU/ml

Urinalysis

The following estimations were performed using the appropriate methodology, as described below:

	Units
Volume	ml
pH - by pH meter	
Specific Gravity (SG) - by refractometry, compared with water to a value of 1000	
Protein - Roche Cobas analyser using modified method of Macart, M. and Gerbaut, L. (1984) *Clin. Chim. Acta.*, 141, 77	mg/dl

Qualitative tests

Total reducing substances (TRS)		Clinitest®
Glucose	)	
Ketones	)	
Bile pigments	)	Multistix®
Urobilinogen	)	
Haem pigments	)	

Clinitest and multistix are diagnostic reagents obtained from Ames Company, Stoke Poges, England and are used as qualitative indicators of analyte concentration. Results are reported according to the following convention:

Tr	=	"trace" of analyte
+	=	"small amount" of analyte
++	=	"moderate amount" of analyte
+++	=	"large amount" of analyte
++++	=	"very large amount" of analyte

For haem. pigments, this degree of differentiation is not possible and the results are reported as negative (0) or positive (+) only.

Microscopy

For microscopic examination, a portion of the urine sample was centrifuged at approximately 1500 g for 10 minutes and the resulting deposit spread on a microscope slide. The deposit was examined for the presence of the following:

Epithelial cells	(E)
Polymorphonuclear leucocytes	(P)
Mononuclear leucocytes	(M)
Erythrocytes	(R)
Organisms	(O)
Renal tubule casts	(C)
Other abnormal constituents	(A)

The presence and approximate frequency of these constituents are reported according to the following convention:

0 = none found in any field examined
1 = a few in some fields examined
2 = a few in all fields examined
3 = many in all fields examined

URINARY INORGANIC FLUORIDE AND ALUMINIUM ANALYSIS

Following routine urinalysis during Week 13, the residual individual samples were pooled for 5 rats of the same sex in each group. During Week 26 (week 13 of withdrawal) urine samples were collected from all withdrawal rats, and pooled for 5 rats of the same sex in each group. All samples were despatched for analysis of inorganic fluoride and aluminium.

Butterworth Laboratories,
54 - 56 Waldegrave Road,
Teddington,
Middlesex,
TW11 8LG.

The samples were received at Butterworth Laboratories on 12 November 1996 (Main study) and 13 February 1997 (Withdrawal).

This ion selective electrode method was developed to determine the concentration of Fluoride in Rat Urine at concentrations between 0.05 mg/l and 100 mg/l in the test solutions. Samples were diluted in a Total Ionic Strengths Adjustment Buffer to release free Fluoride ions and to provide a suitable pH for analysis. In theory, there is no upper limit to the method's range as samples may be diluted many times before analysis.

The fluoride electrode is a chemical sensor in which the detector is a doped single crystal of lanthanum fluoride across which a potential is developed in the presence of fluoride ions. It is normally used with a standard calomel reference electrode and a high impedence millivolt meter or pH meter.

The electrode responds to activity rather than the concentration of the fluoride ions, and to ensure a constant relationship between activity and concentration, samples and calibration standards must be adjusted to a constant ionic strength. They must also be buffered at a suitable pH value to prevent interference by hydroxide ions and also formation of unionised HF under acid conditions. The buffer reagent contains metal-complexing agents (de-complexing agents) to release free fluoride ion from certain metal-fluoride complexes to which the electrode dose not respond. Simple fluoro-silicates are rapidly hydrolysed in water and are detected as fluoride by the electrode.

SACRIFICE

All main study animals were killed following 13 weeks of exposure. Exposure was continued up to the day of sacrifice. Following a 13 week withdrawal period all surviving rats in the withdrawal groups were sacrificed.

Animals were killed by intraperitoneal injection of pentobarbitone sodium and exsanguinated from the brachial blood vessels. Immediately prior to exsanguination, the terminal bodyweight of each animal was recorded.

MACROSCOPIC EXAMINATION AND ORGAN WEIGHTS

All rats were subjected to a detailed macroscopic *post mortem* examination.

The following organs from all animals killed at the scheduled sacrifice were dissected free of fat and weighed:

adrenals	liver	kidneys
lungs	epididymides	testes

PRESERVATION FOR FLUORIDE AND ALUMINIUM LEVELS

Samples of bone and teeth were collected from terminal and withdrawal rats. At necropsy care was taken to avoid contamination of the samples with test substance from the fur.

Bone: A single femur from each rat was removed and scraped clean of adherent muscle and connective tissue. The femurs of all rats in each cage[1] were placed in a labelled plastic container and stored at approximately -20°C prior to despatch for analysis.

Teeth: The lower jaw from each rat was removed cleaned of fur, crushed and the lower incisors removed. The incisors of all rats in each cage[1] were placed in a labelled plastic container and stored at approximately -20°C prior to despatch for analysis.

The samples of bone and teeth for determination of fluoride levels by ion specific electrode and aluminium levels by Inductively coupled Plasma Emission Spectroscopy (ICP) were despatched to:

Butterworth Laboratories Limited
54 - 56 Waldegrave Road
Teddington
Middlesex
TW11 8LG

Tooth and bone samples extracted post mortem were ashed at 500°C. Fluoride was determined in an acid extract of the ground material by Ion Selective (ISE). Aluminium was determined by fusing the ground material in a mix of sodium carbonate and boric acid. After acidification the solution was analysed for aluminium by inductively coupled plasma emission spectrometry (ICP).

Animal 151 female (Group 5 ; High dose Cryolite) was sacrificed during Week 12 of exposure. Samples of bone and teeth were despatched with the other withdrawal samples but was analysed separately.

Animal 84 female (Group 1 ; Air control) died during removal of blood in Week 13. This animal was treated as terminal and samples assigned accordingly.

FIXATION OF TISSUES

Samples, or the whole, of the following organ/tissues, together with any macroscopically abnormal entities were preserved in buffered 10% formalin. The eyes were preserved in Davidson's fixative and the testes and epididymides in Bouin's fixative. The testes were transferred to 70% alcohol after 24 hours. The lungs were infused with fixative prior to immersion.

[b] adrenals
alimentary tract
animal identification mark
[b] aorta (arch and abdominal)
[b] brain (medulla, cerebrum mid-brain and medullary)
[b] caecum
[b] colon
[b] duodenum
[b] eyes
mammary gland
femur and joint (for bone and marrow *in situ*)
[a] gross abnormalities
[b] oesophagus
[b] head (paranasal sinuses, oral cavity, nasopharynx, middle ear, teeth, eyelids, lachrymal gland, Harderian gland and Zymbal's gland)
[b] heart
[b] ileum
[b] jejunum
[b] kidneys
[b] larynx
(2 levels) skin
[b] liver
[c] lungs (all lobes and mainstem bronchi)
[b] lymph nodes (cervical, mesenteric
[c] tracheobronchial
[c] mediastinal)
[b] sternum (bone and marrow)
[b] nasal passages (head for rostral and caudal nasal cavities)
optic nerve
[b] ovaries
[b] pancreas
[b] pharynx
[b] pituitary
prostate
[b] salivary gland
[b] sciatic nerve
seminal vesicle
skeletal muscle (thigh)

spinal column(cervical, thoracic and lumbar)
[b] spleen
[b] stomach (glandular and non-glandular)
[b] rectum
[b] testes (with epididymides)
[b] thymus
[b] thyroids (with parathyroids)
tongue
[b] trachea (including bifurcation)
ureter
[b] urinary bladder
[b] uterus (corpus and cervix)
vagina

MICROSCOPIC PATHOLOGY

Light microscopic examination was performed on 4 μm thick sections, stained with haematoxylin and eosin, of those fixed tissues marked as follows:

- a All rats at termination
- b All rats in Main Groups 1 (Air control), 2 (NaF) and 5 (High dose)
- c All rats in Main and Withdrawal groups

Other macroscopically abnormal tissue(s), and in Groups 3 and 4 tissues, with treatment-related findings in the High dose group were examined.

PREPARATION OF RAT BONE MARROW METAPHASE SLIDES

Twenty four hours after the end of the final exposure (Week 13), rats in the satellite Groups 1 to 5, together with Group 6, were transferred to the Department of Mutagenesis at Huntingdon Life Sciences for processing and preparation of bone marrow slides (4 for each rat). The rats in Group 6 served as a comparative control and were dosed by intraperitoneal injection of cyclophosphamide (20-30 mg/kg) prior to sample removal and processing. Detailed methodology is presented in Appendix 17.

STATISTICAL ANALYSIS

All statistical analyses were carried out separately for males and females.

Food and Water consumption was analysed using cage mean values.

For all other parameters the analyses were carried out using the individual animal as the experimental unit. Bodyweight data were analysed using weight gains. The following sequence of statistical tests was used for bodyweight, organ weight and clinical pathology data.

> If the data consist predominantly of one particular value (relative frequency of the mode exceeded 75%), the proportion of animals with values different from the mode was analysed by appropriate methods. Otherwise:
>
> Bartlett's test(4) was applied to test for heterogeneity of variance between treatments; where significant (at the 1% level) heterogeneity was found, a logarithmic transformation was tried to see if a more stable variance structure could be obtained.
>
> If no significant heterogeneity was detected (or if a satisfactory transformation was found), a one-way analysis of variance was carried out. If significant heterogeneity of variance was present, and could not be removed by a transformation, the Kruskal-Wallis analysis of ranks(5) was used.
>
> Except for pre-exposure data, analyses of variance were followed by Student's *t* test and Williams' test(6) for a dose-related response, although only Williams' test was reported. The Kruskal-Wallis analyses were followed by Shirley's test(7), the non-parametric equivalent of the *t* test and Williams' tests.

Where appropriate, analysis of covariance was used in place of analysis of variance in the above sequence. For organ weight data, the final bodyweight was used as covariate in an attempt to allow for differences in bodyweight which might influence the organ weights.

For microscopic findings Fisher's exact test(8) was employed to detect treatment-related differences.

ARCHIVING

All specimens, raw data and study related documents generated during the course of the study at Huntingdon Life Sciences, together with a copy of the final report were lodged in Huntingdon Life Sciences Archives.

Such specimens and records will be retained for a minimum period of 5 years from the date of issue of the final report. At the end of the 5 year retention period the Sponsor will be contacted and advice sought on the future requirement. Under no circumstances will any item be discarded without the Sponsor's knowledge.

RESULTS

CHAMBER ATMOSPHERE CONDITIONS

Chamber analysed concentration of Cryolite

The data are presented as follows:

Table 1 - Chamber concentrations - daily mean values
Appendix 4 - Chamber concentrations - individual values

The data are summarised below:

Group	Study mean concentration (mg/m³)	
	Target	Analysed
2 (NaF)	6.0	5.7
3 (Low dose Cryolite)	0.2	0.21
4 (Inter dose Cryolite)	1.0	1.04
5 (High dose Cryolite)	5.0	4.6

The analysed concentrations were in good agreement with the target concentrations.

Particle size distribution

The data are presented as follows:

Figure 6 - Particle size distribution of Cryolite
Table 2 - Particle size distribution - individual and sample stage values

The data are summarised below:

Group	Study mean		
	MMAD	(σg)	%<7µm
2 (NaF)	3.1	(2.12)	86
3 (Low dose Cryolite)	1.9	(2.92)	89
4 (Inter dose Cryolite)	2.6	(2.45)	86
5 (High dose Cryolite)	2.8	(2.30)	86

MMAD Mass median aerodynamic diameter
σg Geometric standard deviation

The size distribution of airborne particles at each level of exposure were similar. It is considered that particles below 7 µm aerodynamic diameter are respirable to the rat.

Nominal concentration of Cryolite

The data are presented as follows:

Table 3 - Calculation of nominal concentration

The data are summarised below:

Group	Analysed conc (mg/m^3)	Study mean nominal conc (mg/m^3)	A/N (%)
2 (NaF)	5.7	47.0	12
3 (Low dose Cryolite)	0.21	0.85	25
4 (Inter dose Cryolite)	1.04	4.5	23
5 (High dose Cryolite)	4.6	23.3	20

$$A/N \frac{\text{Analysed concentration}}{\text{Nominal concentration}} \times 100$$

Differences between analysed and nominal concentrations of particulate reflect losses due to impaction and deposition within the exposure system and are considered not unusual for the exposure system employed. The lower efficiency for NaF may reflect different physical properties of this material when generated (eg higher static charge resulting in higher losses in the system).

Temperature

The data are presented as follows:

Table 4 - Chamber temperature - exposure mean values

The data are summarised below:

Group	Study mean Temperature (°C)
1 (Air control)	23.4
2 (NaF)	22.9
3 (Low dose Cryolite)	21.1
4 (Inter dose Cryolite)	21.5
5 (High dose Cryolite)	22.3

The small differences between groups are considered not to have affected the outcome of the study.

CLINICAL OBSERVATIONS

Mortality

Rat 106F, Group 2 (NaF; terminal) was found dead in its restraining tube at the end of the exposure on 13 September 1996 (Week 6). This death was considered incidental and not related to exposure.

Rat 84F, Group 1 (Air control; terminal) died during removal of blood on 5 November 1996 (Week 13).

Rat 151F, Group 5 (High dose Cryolite; withdrawal) was sacrificed on humane grounds on 1 November 1996 (Week 12).

Clinical signs

The data are presented as follows:

Appendix 5 - Individual clinical signs

No treatment-related clinical signs were seen either during or between exposures.

Hairloss, abrasions, brown staining on head/body and red staining around the eyes were seen between exposures. These were considered incidental and not treatment-related.

Following exposure on 10 October 1996 (Week 10) rat 66 male Group 5 (High dose Cryolite; Terminal) was accidentally returned to an incorrect cage; Cage 31 female Group 5 (High dose Cryolite; Withdrawal). When the rat was discovered in the wrong cage the following morning 2 vaginal plugs were present in the tray beneath the cage. Exposure of the rats in Cage 31 female continued. Body weight was monitored daily for all rats in the affected cage in order to determine any definite pregnancy. In addition each rat was palpated at intervals over the following 2 weeks.

Eventually it was determined that only rat 151 female was actually (or remained) pregnant. Accordingly, in order for birth to take place 151 female was not exposed on 31 October (Week 13), Day 21 of pregnancy. Spontaneous delivery had not occurred by the following morning (1 November 1996), and the condition of this rat had deteriorated to such an extent that survival was doubtful. Accordingly the rat was sent for necropsy for humane reasons. At necropsy bone and tooth samples were obtained together with a full range of tissues preserved.

Bodyweight

The data are presented as follows:

Figure 7 and 8	-	group mean values
Table 5	-	group mean values
Appendix 6	-	individual values

No treatment-related effects were seen.

Food consumption

The data are presented as follows:

Table 6	-	group mean values
Appendix 7	-	cage mean values

No treatment-related effects were seen.

Water consumption

The data are presented as follows:

Table 7 - group mean values
Appendix 8 - individual cage mean values

No treatment-related effects were seen.

Ophthalmic examination

The data are presented as follows:

Appendix 9 - individual findings

No treatment-related effects were seen.

LABORATORY INVESTIGATIONS

Haematology

The data are presented as follows:

Table 8 - group mean values
Appendix 10 - individual values

No treatment-related differences were seen.

Increased total and differential white cell counts were evident in groups exposed to Cryolite, especially for female rats in Group 5 (High dose). However, the differences are within the historical range of background data for rats of the age and strain used.

Other differences of statistical significance were confined to a single sex, within the historical range of background data for rats of the age and strain used, and were considered not to be of toxicological significance.

Biochemistry

The data are presented as follows:

Table 9 - group mean values
Appendix 11 - individual values

No treatment-related differences were evident.

Increased electrolyte concentrations were evident in groups exposed to Cryolite, especially for female rats in Group 5 (High dose). However, the differences are within the historical range of background data for rats of the age and strain used.

Other differences of statistical significance were confined to a single sex, within the historical range of background data for rats of the age and strain used, and were considered not to be of toxicological significance.

Other differences achieving statistical significance were small and/or inconsistent between the sexes and considered not of toxicological significance.

Urinalysis

The data are presented as follows:

Table 10 - group mean values
Appendix 12 - individual values

No treatment-related differences were seen.

Urinary inorganic fluoride and aluminium

The data are presented as follows:

Table 11 - group mean values
Appendices 13 and 14 - pooled values

Group	Fluoride (mg/litre)			
	Terminal		Withdrawal	
	Male	Female	Male	Female
1 (Air control)	1.5	1.35	1.05	1.08
2 (NAF)	6.5	7.25	1.5	1.3
3 (Low dose control)	1.25	1.4	-	
4 (Inter dose control)	1.4	1.5	-	
5 (High dose control)	3.8	4.6	0.95	1.2

Increased fluoride concentrations were evident in pooled urine samples from Groups 2 (NaF) and 5 (High dose Cryolite). The increases were slightly greater in female rats. No evidence of fluoride increases were seen at the Low and Intermediate Cryolite levels.

After 13 week recovery the fluoride levels in Groups 1 (Air control), 2 (NaF) and 5 (High dose Cryolite) were similar and considered normal.

Group	Aluminium (% w/w)			
	Terminal		Withdrawal	
	Male	Female	Male	Female
1 (Air control)	1.5	<0.6	<0.5	<0.5
2 (NAF)	0.5	<0.6	<0.5	0.24
3 (Low dose control)	<0.5	2.5	-	
4 (Inter dose control)	0.6	1.1	-	
5 (High dose control)	1.8	2.6	<0.5	<0.5

Aluminium concentrations were increased in both sexes in Group 5 (High dose Cryolite). Increased concentrations were evident for female rats in Group 3 (Low dose Cryolite) and in both sexes in Group 4 (Intermediate dose Cryolite). However, a dose relationship was not evident.

After 13 weeks withdrawal all groups were similar and a degree of recovery was considered to have occurred. An isolated high value in Group 2 (NaF) at withdrawal was considered unlikely to be related to treatment.

TERMINAL STUDIES

Bone and teeth analysis for fluoride and aluminium

The data are presented as follows:

Tables 12 and 13	-	group mean values
Appendices 13 and 14	-	pooled values

Group	Bone fluoride (% w/w)			
	Terminal		Withdrawal	
	Male	Female	Male	Female
1 (Air control)	0.026	0.032	0.027	0.037
2 (NAF)	0.057	0.079	0.054	0.073
3 (Low dose control)	0.022	0.030	-	-
4 (Inter dose control)	0.018	0.028	-	-
5 (High dose control)	0.041	0.063	0.039	0.064

Fluoride concentrations in bone were increased in rats of both sexes in Groups 2 (NaF) and 5 (High dose Cryolite) at termination of exposures. The concentration of fluoride in bone after 13 weeks withdrawal remained similar to terminal concentrations and no evidence for recovery was seen.

Group	Tooth fluoride (% w/w)			
	Terminal		Withdrawal	
	Male	Female	Male	Female
1 (Air control)	0.015	0.018	0.013	0.016
2 (NAF)	0.020	0.018	0.011	0.013
3 (Low dose control)	0.017	0.013	-	-
4 (Inter dose control)	0.017	0.018	-	-
5 (High dose control)	0.017	0.023	0.010	0.013

Increased fluoride concentration in tooth samples was also seen in Groups 2 (NaF) and 5 (High dose Cryolite) at termination of exposures. However, the differences from control were slight, with the values for male rats in Group 5 (High dose Cryolite) being similar to controls. After 13 weeks of withdrawal from exposure the concentration of fluoride in all groups was similar, and a degree of recovery was considered to have taken place.

At an equivalent level of Fluoride exposure it would appear from the results obtained in this study that release and accumulation of fluoride ion was greater from sodium fluoride than Cryolite.

Aluminium concentrations for pooled terminal and withdrawal samples of bone and teeth in all groups, were below the level of quantification of the method of analysis employed, <0.01% w/w. No treatment-related effects were detectable.

Macroscopic pathology

The data are presented as follows:

Tables 14 and 15	-	incidence summary
Appendix 18	-	individual findings

No treatment-related changes were seen at necropsy.

Macroscopic findings at necropsy of Rat 151F (Group 5 - withdrawal) sacrificed due to poor condition 1 November 1996 were as follows:

Fur: red/brown staining in the perinatal, periorbital and cranial regions;
GI tract: minimal contents
Stomach-corpus mucosa: haemorrhagic depression (3 mm)
Adrenals: minimally enlarged (0.1134 g)
Uterus: contained 17 foetuses
Lumber Lymph nodes: minimal enlargement

Organ weights

The data are presented as follows:

Tables 16 and 17	-	group mean values
Appendices 15 and 16	-	individual values

A statistically significant increase in the lung weights of both sexes of rats in Group 5 (High dose Cryolite) was present at termination, compared with Air control rats, $p < 0.01$.

A similar effect in rats exposed to NaF at an equivalent level of fluoride was not seen.

A slight increase in the lung weights of rats in Group 5 (High dose Cryolite) was evident at withdrawal achieving statistical significance in male rats only, $p<0.01$ using Students 't' test.

No other treatment-related differences were seen.

Microscopic pathology

The data are presented as follows:

Tables 18 to 20	-	incidence summary
Appendix 18	-	individual findings

Microscopic changes seen in Rat 151F (Group 5 - withdrawal) sacrificed due to poor condition 1 November 1996 were as follows:

Lungs: alveolitis with trace intestinal thickening of alveolar duct walls
Stomach: trace erosion of glandular epithelium

The following comments are made in summary:

Treatment related changes

Lungs

Comparative control/NaF treated animals - Aggregations of alveolar macrophages were seen in a proportion of animals from the sodium fluoride treated group. These aggregations were focal and seen either distributed throughout the lung parenchyma or localised around alveolar ducts.

Cryolite treated animals - In the majority of animals from the High dose group treatment-related changes in the lungs comprised varying degrees of macrophage aggregation around alveolar ducts and alveolitis with thickening of alveolar duct walls. In addition a majority of animals possessed aggregations of alveolar macrophages which contained brown pigmented material. These changes were statistically significant when compared to the incidence in control group animals.

Other changes seen that were considered to be associated with treatment in some animals included perivascular inflammatory infiltration. Increased collagen in alveolar duct walls and extension of bronchiolar epithelium into alveolar ducts.

Alveolitis with interstitial thickening of the alveolar duct walls was also present in a proportion of Intermediate dose rats and was considered to be associated with treatment. The change in this group was less severe than that seen in the High dose group animals that were affected.

1 mg/m³

Rat 151 female Group 5 (High dose Cryolite) - withdrawal) sacrificed on humane grounds during Week 12 exhibited alveolitis with trace interstitial thickening of alveolar duct walls of the lungs, and trace erosion of the glandular epithelial of the stomach.

8 mg/m³

TERMINAL KILL	Males					Females				
	Con	NaF	Low	Int	High	Con	NaF	Low	Int	High
Macrophages around alveolar ducts	0	0	0	0	9**	0	0	0	0	6*
Aggregations of macrophages	1	4	2	0	0	0	1	0	0	0
Increased collagen in alveolar ducts	0	0	0	0	2	0	0	0	0	0
Extension of bronchiolar epithelium into alveolar ducts	0	0	0	0	3	0	0	0	0	0
Alveolitis with interstitial thickening of alveolar duct walls	0	0	0	5*	10**	0	1	0	3	8**
Brown pigment in macrophages	0	0	0	1	9**	0	0	0	1	8**
Macrophages in BALT containing brown pigment	0	0	0	0	5*	0	0	0	0	1
Number of lungs examined	10	10	10	10	10	9	9	10	10	10

* $p<0.05$ ** $p<0.01$ with Fisher's Exact Test

After a period of recovery/withdrawal the above treatment related changes were seen to have resolved with the exception of the presence of small foci of brown pigmented alveolar macrophages in occasional animals, although these were present to a lesser degree than at termination of the dosing period.

A background incidence of low-grade respiratory inflammation was present in both control and treated animals after the conclusion of the recovery period. This was not associated with treatment.

RECOVERY KILL	Males					Females				
	Con	NaF	Low	Int	High	Con	NaF	Low	Int	High
Pneumonitis	3	9**			6	2	3			3
Perivascular inflammatory infiltration	7	10			9	3	7			8*
Alveolitis with interstitial thickening of alveolar duct walls	0	0			1	0	0			1
Brown pigment in macrophages	0	0			4*	0	0			4*
Number of lungs examined	10	10			10	10	10			9

* $p<0.05$ ** $p<0.01$ with Fisher's Exact Test

Larynx

Comparative control/NaF treated animals - Ventral epithelial hyperplasia and subepithelial inflammation was seen in a majority of animals from the positive control group at termination. Similar changes were not seen in air control group animals.

Cryolite treated animals - No treatment related laryngeal changes were seen in Cryolite treated animals.

TERMINAL KILL	Males					Females				
	Con	NaF	Low	Int	High	Con	NaF	Low	Int	High
Larynx										
Ventral epithelial hyperplasia	0	8**	0	0	0	0	8**	0	0	0
Ventral subepithelial inflammation	0	5*	0	0	0	0	4*	0	0	0
Number of larynges examined	10	10	0	0	10	9	9	0	0	10

* $p<0.05$ ** $p<0.01$ with Fisher's Exact Test

The larynx was not examined for animals of the recovery kill groups.

Lymph nodes

Comparative control/NaF treated animals - No treatment related changes were seen the lymph nodes of sodium fluoride treated animals.

Cryolite treated animals - Brown pigment containing macrophages distinct from siderocytes were seen in small numbers in the tracheobronchial and mediastinal lymph nodes at termination.

TERMINAL KILL	Males					Females				
	Con	NaF	Low	Int	High	Con	NaF	Low	Int	High
Tracheobronchial lymph nodes										
Macrophages with brown pigment	0	0	0	1	7**	0	0	0	0	6*
Mediastinal lymph nodes										
Macrophages with brown pigment	0	0	0	0	4*	0	0	0	0	3
Number of rats examined	10	10	10	10	10	9	9	10	10	10

* $p<0.05$ ** $p<0.01$ with Fisher's Exact Test

After the conclusion of the recovery/withdrawal period brown pigmented macrophages were no longer discernible in the affected lymph nodes.

Incidental changes

All other microscopic findings were considered to be incidental and of no toxicological importance.

No changes were seen in the nasal passages of animals from either the sodium fluoride or test material exposed groups.

Conclusions

Pulmonary inflammatory lesions associated with exposure to Cryolite were seen in a majority of animals in the High dose group and, to a lesser degree, in some animals from the Intermediate group.

No treatment-related changes were seen in animals from the Low dose cryolite group.

The treatment related changes had, with the exception of the presence of small foci of brown pigmented alveolar macrophages, resolved after the conclusion of the recovery/withdrawal period.

Animals from the sodium fluoride (comparative control) group exhibited laryngeal epithelial hyperplastic and inflammatory changes. Laryngeal changes were not seen in animals receiving Cryolite.

The Low dose group (0.21 mg/m^3) was considered to be the no-effect level for exposure to Cryolite in this study.

DISCUSSION

In this study, groups of rats were exposed snout only to particulate aerosols of Cryolite or sodium fluoride, 6 hours a day, 5 days a week for 13 consecutive weeks. Withdrawal rats in the air control, sodium fluoride NaF and High dose Cryolite groups were retained unexposed for 13 weeks following the last exposure.

The study mean analysed concentration of the aerosol of sodium fluoride was 5.7 mg/m^3, and of Cryolite were 0.21, 1.04 and 4.6 mg/m^3 in air.

No effects of treatment were evident in clinical signs, bodyweight gain, food or water consumption. Haematological, biochemical and urinalysis parameters did not indicate findings considered of toxicological significance. Treatment-related macroscopic findings were not seen at terminal or withdrawal necropsy.

Following 13 Weeks of dosing, there was a treatment related statistically significant increase in the lung weight values for Group 5 (High dose Cryolite) rats of both sexes. A similar but less obvious effect was present following 13 weeks of withdrawal; Group 5 male rats remaining statistically significantly higher compared with controls, but a similar difference evident in female High dose rats did not attain statistical significance.

Increased fluoride concentrations in urine, bones and teeth were evident for rats in Group 2 (NaF) and 5 (High dose Cryolite). After withdrawal from exposure for 13 weeks urinary fluoride was similar between groups, but bone concentrations remained similar to values at the end of 13 weeks exposure.

Aluminium concentrations in the urine were increased in groups exposed to Cryolite, being most obvious at the High dose level. Increases in Low and Intermediate group samples were not dose-related. Analysed values for aluminium in bone and teeth were below the limit of detection for the method of analysis used, and there was no evidence for accumulation of aluminium in rats during the 13 week period of exposures.

At termination, histopathology considered related to treatment with the comparative control substance, sodium fluoride, was confined to focal aggregations of alveolar macrophages in the lung parenchyma and around the alveolar ducts, together with ventral epithelial hyperplasia and subepithelial inflammation of the larynx.

In rats at the High dose of Cryolite, lung histopathology also revealed macrophage aggregations but their distribution was in the alveolar ducts. In addition, alveolitis, thickening of the alveolar duct walls together with macrophages containing brown pigment were seen. Macrophages containing brown pigmented material were present in the tracheobronchial and mediastinal lymph nodes of High dose rats. A proportion of High dose rats had perivascular inflammatory infiltration with increased collagen in the alveolar duct walls, with extension of bronchiolar epithelium into the alveolar ducts. At the Intermediate dose of Cryolite, a proportion of rats had interstitial thickening of the alveolar duct walls. The lung changes in Cryolite exposed rats are typical of a non-specific reaction over time to a particulate with irritant properties, and attempts at clearance of deposited material via the lung macrophage/lymph node route.

The response of the respiratory tract to inhalation exposure to sodium fluoride differed from the response to exposure to Cryolite at a similar concentration and particle size. In rats exposed to sodium fluoride lesions were seen in the larynx, where as in rats exposed to Cryolite lesions were in the lungs. The reasons for this difference is not known but may be related to the relative solubility of Cryolite and Sodium fluoride. Sodium fluoride is more soluble than Cryolite and may not remain in the lungs in particulate form for a period of time sufficient to cause the degree of response seen with Cryolite.

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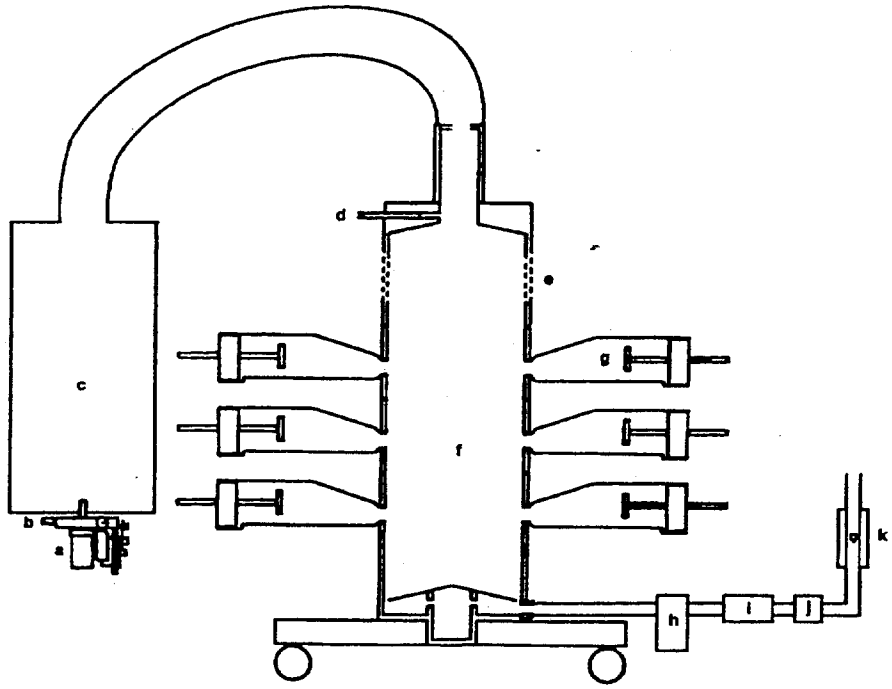
FIGURE 1

Schematic diagram of the inhalation exposure system

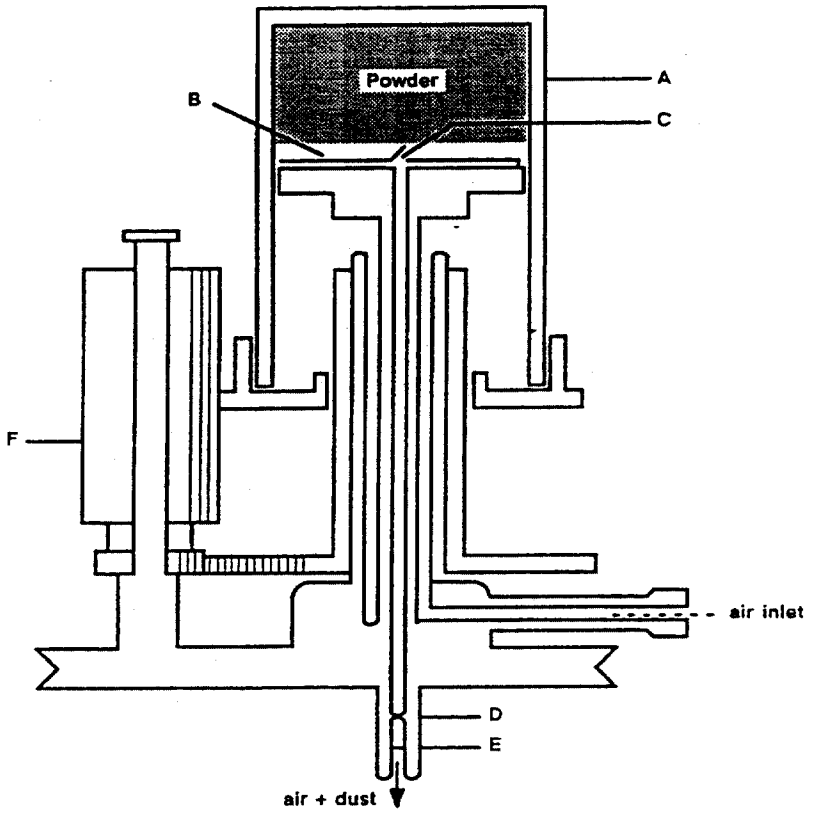


Key

a	Wright dust feed mechanism	f	Inhalation chamber
b	Air supply (10 l/minute; 5 litre/minute Group 3)	g	Restraining tube
c	Elutriation canister	h	Temp/RH probe
d	Balance air inlet (49l/minute)	i	Glass wool filter
e	Observation port (1 of 4)	j	Silica gel drier
		k	Exhaust rotameter (60 l/minute)

FIGURE 2

Wright dust feed mechanism



A	Canister
B	Scraper
C	Outlet tube
D	Jet
E	Baffle plate
F	Drive gear

FIGURE 3

Rodent nasal turbinates - sectioning levels

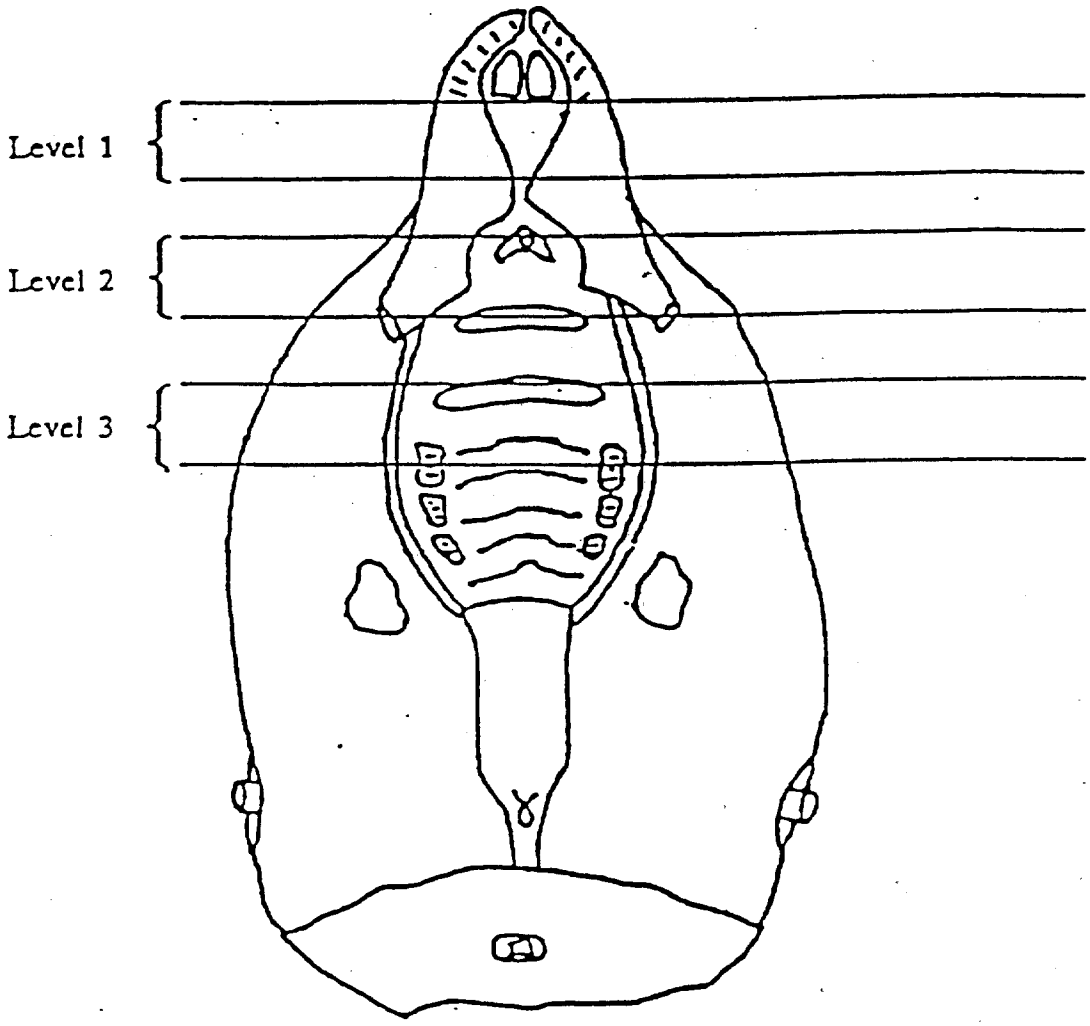


FIGURE 4

Levels of larynx processed for histology

Larynx

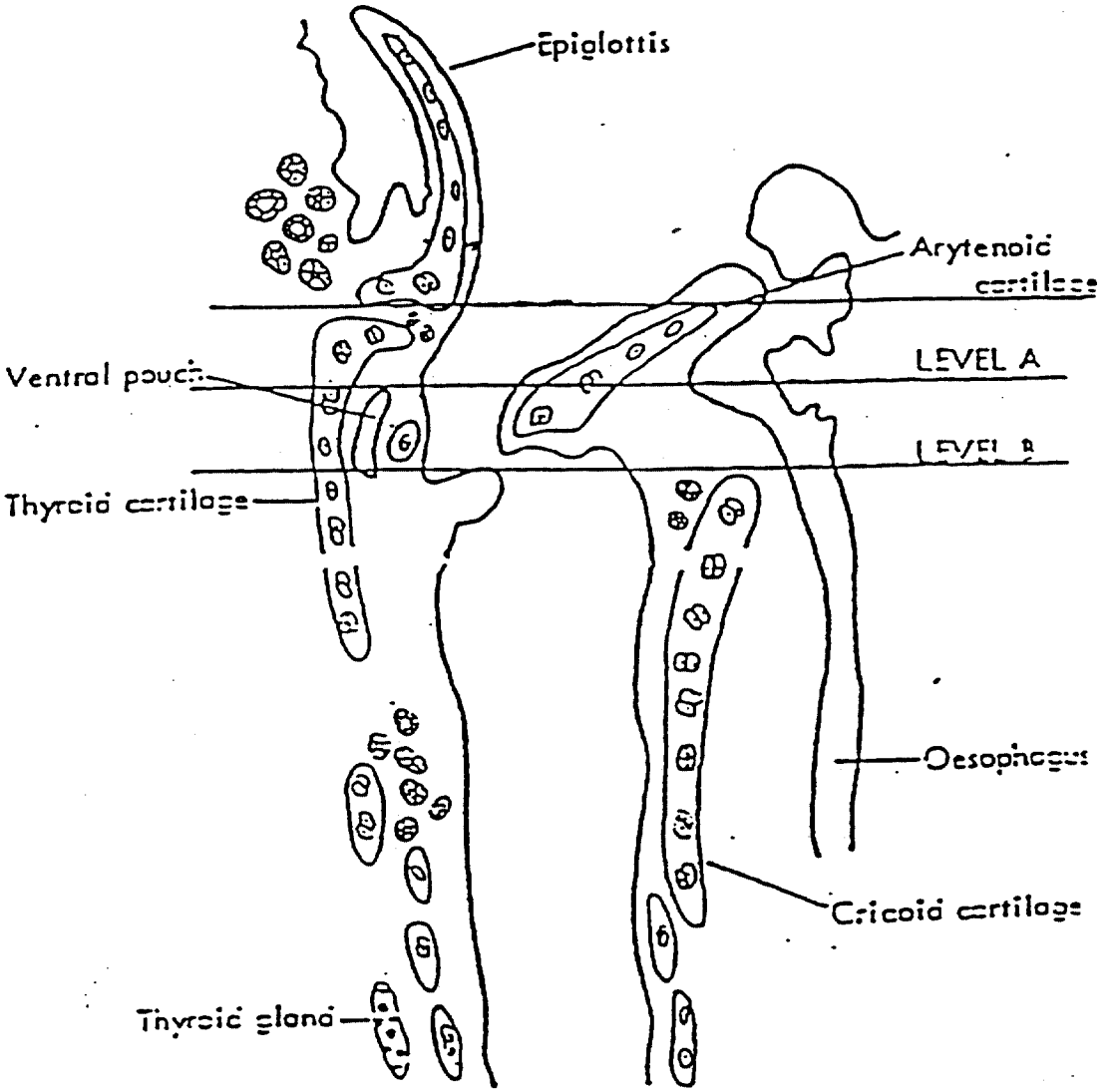


FIGURE 5

Rodent larynx - sectioning for inhalation studies

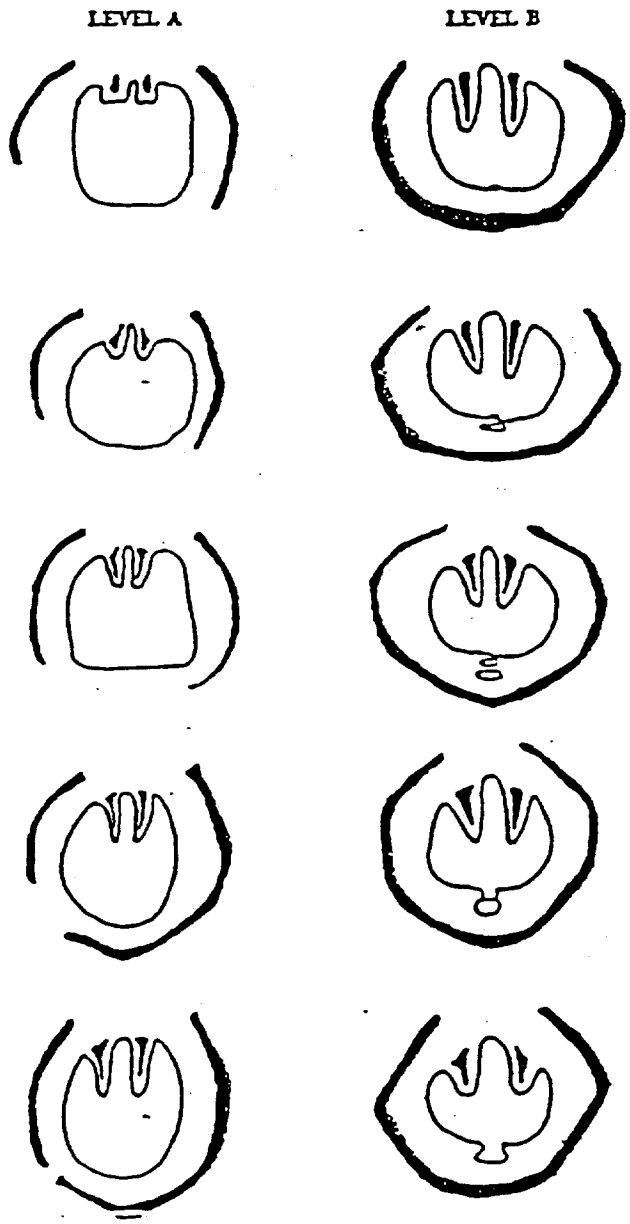


FIGURE 6

Particle size distribution of Cryolite

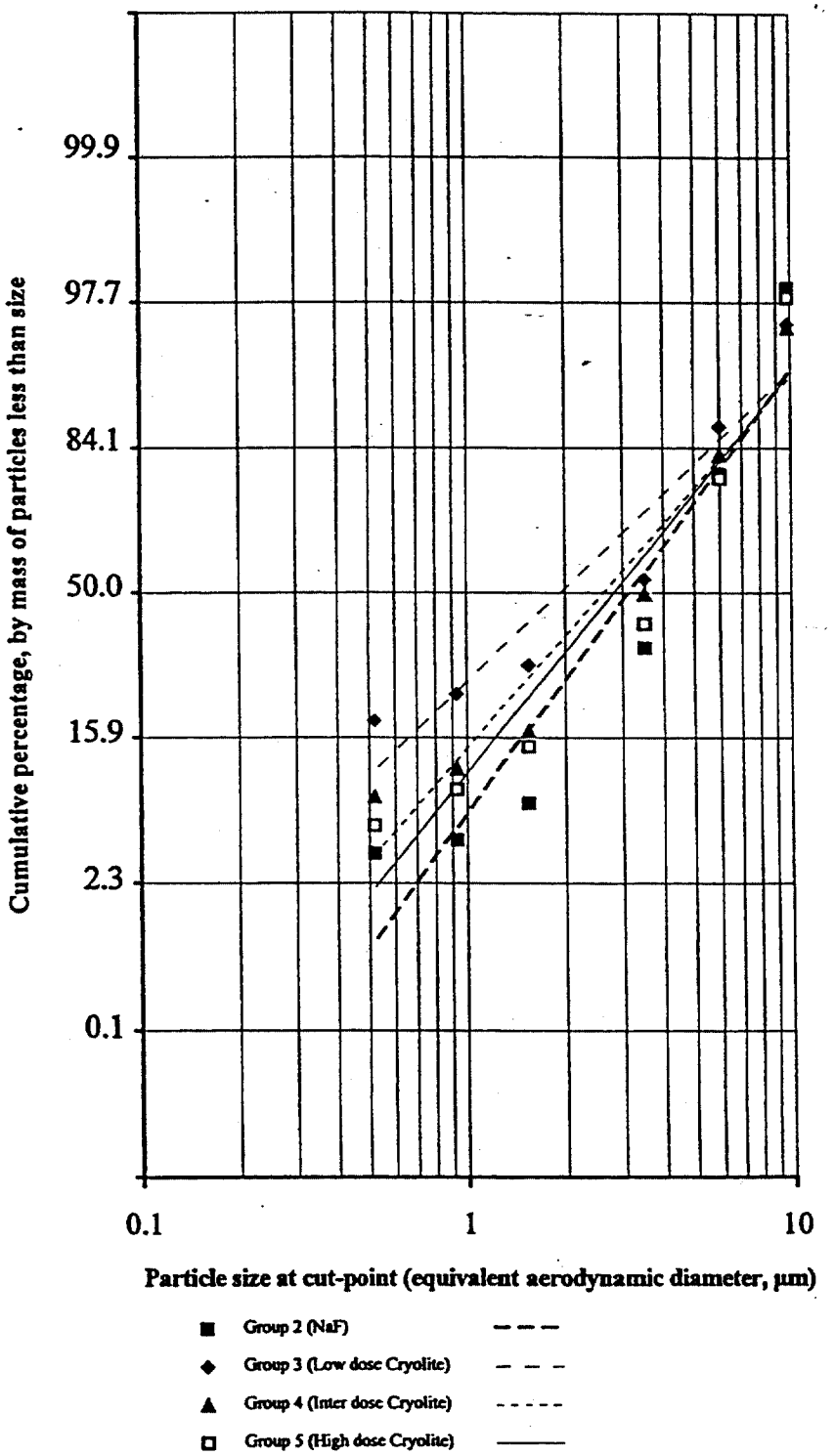


FIGURE 7

Bodyweights- group mean values - 13 week exposure period

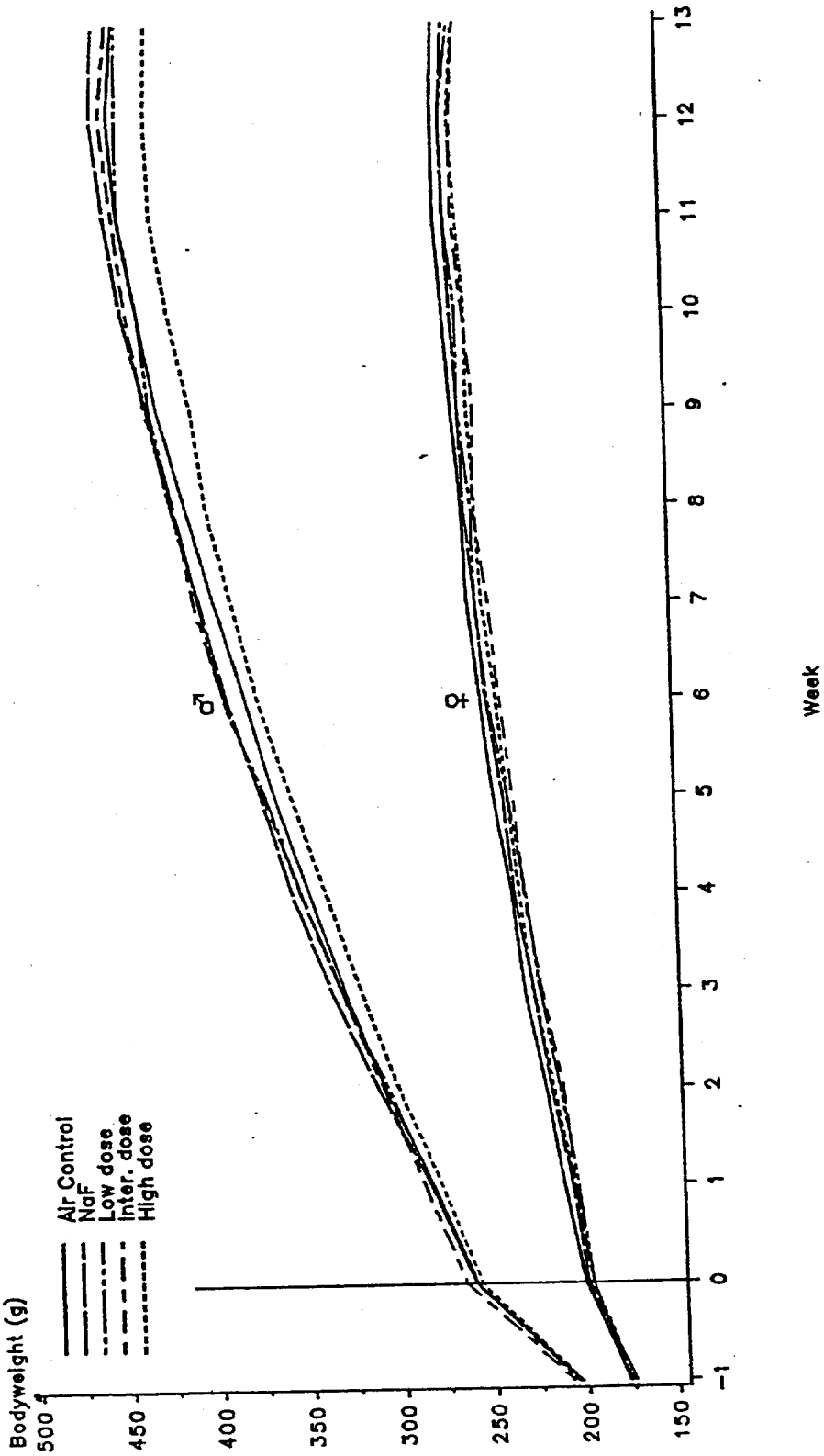


FIGURE 8

Bodyweights- group mean values - withdrawal rats only

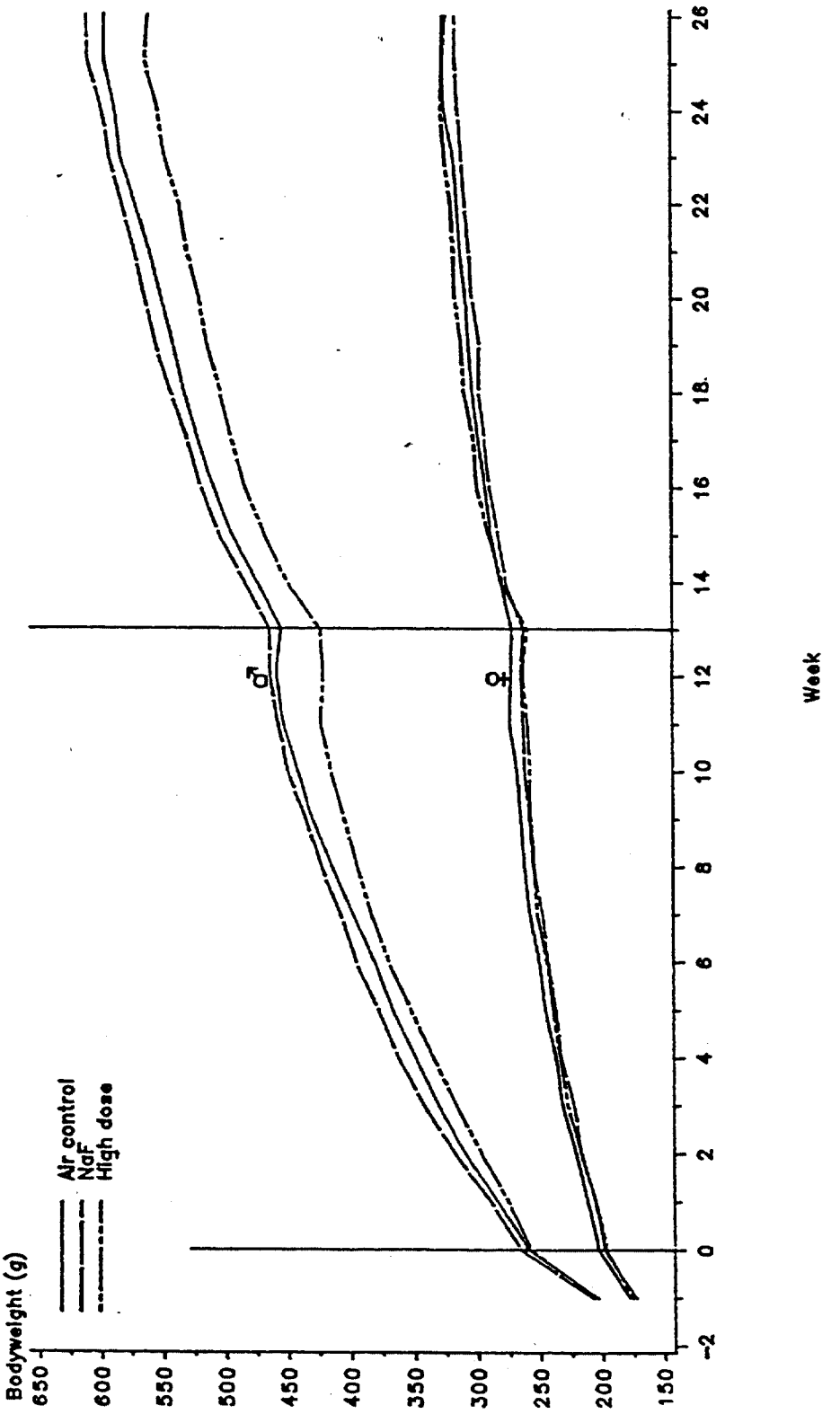


TABLE 1

Chamber concentration - daily mean values (mg/m^3)

Exposure	Group			
	2 Na F	3 Low dose Cryolite	4 Inter dose Cryolite	5 High dose Cryolite
1	6.0	0.18	1.14	5.7
2	5.9	0.25	0.83	3.6
3	5.6	0.24	1.31	4.6
4	5.9	0.10	1.12	4.3
5	5.6	0.34	1.17	5.1
6	6.0	0.37	0.96	3.7
7	5.6	0.31	1.19	4.6
8	7.1	0.29	1.68	7.9
9	5.7	0.16	0.85	2.5
10	6.2	0.48	1.28	4.9
11	6.1	0.20	1.24	4.4
12	6.0	0.20	1.33	4.7
13	5.3	0.22	1.26	6.1
14	5.3	0.21	1.34	4.8
15	5.0	0.22	1.26	4.9
16	5.5	0.22	1.27	4.5
17	6.4	0.23	1.27	4.5
18	5.8	0.24	1.30	4.6
19	5.8	0.23	1.26	4.4
20	5.3	0.23	1.25	4.4
21	5.6	0.20	1.15	4.1
22	5.7	0.22	1.28	4.5
23	2.3	0.25	1.31	4.8
24	5.1	0.22	1.11	4.1
25	3.1	0.21	1.25	4.3
26	4.4	0.20	1.17	4.2
27	5.7	0.27	1.24	4.8
28	4.7	0.22	1.20	4.1
29	6.3	0.26	1.23	4.8
30	5.1	0.23	1.08	3.8
31	5.1	0.19	0.88	4.4
32	5.9	0.15	0.99	4.6

TABLE 1

Chamber concentration - daily mean values (mg/m^3)

Exposure	Group			
	2 Na F	3 Low dose Cryolite	4 Inter dose Cryolite	5 High dose Cryolite
33	5.7	0.16	0.83	4.2
34	6.1	0.21	0.74	4.2
35	5.8	0.18	1.10	4.5
36	5.9	0.19	0.88	4.3
37	5.9	0.21	0.95	4.8
38	5.9	0.08	0.89	4.9
39	6.1	0.22	1.05	4.8
40	6.1	0.20	0.85	4.7
41	6.4	0.22	0.91	4.6
42	5.5	0.14	1.15	4.2
43	5.8	0.18	0.55	4.0
44	5.9	0.22	0.88	4.5
45	6.3	0.17	0.74	4.5
46	6.3	0.23	0.87	4.4
47	6.5	0.19	0.89	4.5
48	6.2	0.20	0.84	4.4
49	5.6	0.18	0.85	4.5
50	5.4	0.20	0.88	4.6
51	5.2	0.14	0.83	4.5
52	6.0	0.10	0.85	4.0
53	5.2	0.17	0.76	4.7
54	6.8	0.18	1.00	4.9
55	5.9	0.18	0.99	4.2
56	6.0	0.20	1.05	5.2
57	5.6	0.25	0.87	4.5
58	5.5	0.16	0.83	4.1
59	5.3	0.23	1.14	5.5
60	5.4	0.17	0.84	4.3
61	5.7	0.21	0.99	5.4
62	5.7	0.21	0.85	4.5
63	5.7	0.20	0.89	4.5
64	5.4	0.20	0.87	5.3
65	6.1	0.19	1.04	5.3
66	6.1	0.23	0.90	5.3
Mean	5.7	0.21	1.04	4.6
sd	0.70	0.057	0.206	0.66

sd Standard deviation

TABLE 2

Particle size distribution - individual and sample stage values

Group	Week	% of total collected on each stage							Particle size		
		3	4	5	6	7	8	Filter	MMAD	σg	% < 7μm
2	1	0.0	11.1	61.5	25.5	1.4	0.0	0.5	3.1	1.67	94
(NaF)	2	2.0	19.2	52.3	21.9	4.6	0.0	0.0	4.0	1.63	88
	3	0.0	18.3	41.5	32.3	3.1	0.4	4.4	2.6	1.91	94
	4	1.1	31.0	41.9	22.9	1.4	0.0	1.8	3.6	1.98	83
	5	2.1	22.9	44.7	27.7	0.0	0.0	2.7	3.6	2.06	82
	6	0.0	31.8	42.9	20.9	0.7	0.3	3.4	3.0	1.93	90
	7	0.0	13.4	49.2	29.1	2.8	0.0	5.6	2.5	1.92	94
	8	0.0	13.3	39.9	36.7	2.8	1.4	6.0	2.4	1.94	95
	9	4.1	16.4	43.5	32.3	2.2	0.0	1.5	3.7	2.02	82
	10	2.6	15.0	36.8	33.1	3.8	3.0	5.6	2.8	2.29	87
	11	2.9	17.5	36.0	31.6	3.6	2.2	6.2	2.9	2.35	85
	12	3.9	22.0	31.0	36.6	3.4	0.9	2.2	3.5	2.14	82
	13	3.8	14.4	53.8	10.6	8.3	2.3	6.8	3.0	2.49	83
	Mean	1.7	18.9	44.2	27.8	2.9	0.8	3.6	3.1	2.12	86

MMAD Mass median aerodynamic diameter
σg standard deviation

Collection characteristics

Marple personal cascade impactor cut-points at 2 l/minute

Impactor stage	3.00	4.00	5.00	6.00	7.00	8.00	Filter
Cut-point* (μm)	9.80	6.00	3.50	1.55	0.93	0.52	0.00

* Aerodynamic equivalent particle for spherical particles of unit mass density in air at 25°C and 760 mmHg

Calculation of MMAD, σg and inhalable fraction (% < 7μm)

By linear regression - probit of cumulative percentage , by mass , of particles smaller than cut-point of each stage versus the logarithm of the cut-point of each stage.

TABLE 2

Particle size distribution - individual and sample stage values

Group	Week	% of total collected on each stage							Particle size		
		3	4	5	6	7	8	Filter	MMAD	σg	% < 7μm
3	1	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
(Low dose	2	2.9	8.6	50.0	28.6	2.9	2.9	4.3	2.8	2.17	88
Cryolite)	3	0.0	8.3	31.3	29.2	6.3	2.1	22.9	1.6	2.26	97
	4	4.5	7.6	30.3	21.2	10.6	6.1	19.7	1.8	3.10	88
	5	0.0	7.0	41.9	16.3	4.7	2.3	27.9	1.5	2.37	96
	6	0.0	7.3	46.3	17.1	4.9	0.0	24.4	1.6	2.29	96
	7	8.0	12.0	28.0	24.0	0.0	8.0	20.0	2.2	3.71	81
	8	9.1	7.3	29.1	16.4	9.1	7.3	21.8	1.9	3.88	83
	9	2.4	10.8	25.3	22.9	7.2	3.6	27.7	1.6	3.22	90
	10	4.9	9.8	36.6	23.2	4.9	12.2	8.5	2.4	2.69	86
	11	0.0	7.7	26.9	26.9	15.4	3.8	19.2	1.6	2.21	97
	12	0.0	7.3	43.9	26.8	2.4	7.3	12.2	1.9	2.09	96
	13	5.7	18.9	15.1	22.6	11.3	9.4	17.0	2.0	3.44	84
	Mean	3.1	9.4	33.7	22.9	6.6	5.4	18.8	1.9	2.92	89

MMAD Mass median aerodynamic diameter
σg standard deviation

Collection characteristics

Marple personal cascade impactor cut-points at 2 l/minute

Impactor stage	3.00	4.00	5.00	6.00	7.00	8.00	Filter
Cut-point* (μm)	9.80	6.00	3.50	1.55	0.93	0.52	0.00

* Aerodynamic equivalent particle for spherical particles of unit mass density in air at 25°C and 760mmHg

Calculation of MMAD, σg and inhalable fraction (% < 7μm)

By linear regression - probit of cumulative percentage , by mass , of particles smaller than cut-point of each stage versus the logarithm of the cut-point of each stage.

TABLE 2

Particle size distribution - individual and sample stage values

Group	Week	% of total collected on each stage							Particle size		
		3	4	5	6	7	8	Filter	MMAD	σg	% < 7μm
4	1	2.0	11.2	35.7	42.9	6.1	1.0	1.0	3.1	1.90	90
(Inter dose	2	1.5	13.0	33.6	33.6	5.3	3.1	9.9	2.3	2.35	90
Cryolite)	3	0.8	21.8	41.4	24.8	3.8	1.5	6.0	2.8	2.17	89
	4	1.4	19.6	33.1	23.6	7.4	4.7	10.1	2.4	2.48	88
	5	1.5	15.8	35.3	28.6	3.0	1.5	14.3	2.3	2.53	89
	6	4.9	13.6	27.7	25.5	6.5	7.1	14.7	2.2	3.01	85
	7	3.2	11.7	37.2	34.0	2.1	4.3	7.4	2.6	2.40	87
	8	2.9	2.9	32.4	50.0	1.5	0.0	10.3	2.3	2.25	92
	9	0.0	14.6	38.2	38.2	6.7	2.2	0.0	2.9	1.63	96
	10	0.9	16.4	35.5	35.5	9.1	0.0	2.7	2.8	1.96	91
	11	14.7	17.3	24.0	28.0	8.7	5.3	2.0	3.8	2.60	74
	12	1.1	12.2	33.3	30.0	2.2	5.6	15.6	2.0	2.51	91
	13	8.3	9.3	25.9	29.6	12.0	6.5	8.3	2.6	2.80	84
	Mean	3.3	13.8	33.3	32.6	5.7	3.3	7.9	2.6	2.45	86

MMAD Mass median aerodynamic diameter
σg standard deviation

Collection characteristics

Marple personal cascade impactor cut-points at 2 l/minute

Impactor stage	3.00	4.00	5.00	6.00	7.00	8.00	Filter
Cut-point* (μm)	9.80	6.00	3.50	1.55	0.93	0.52	0.00

* Aerodynamic equivalent particle for spherical particles of unit mass density in air at 25°C and 760mmHg

Calculation of MMAD, σg and inhalable fraction (% < 7μm)

By linear regression - probit of cumulative percentage , by mass , of particles smaller than cut-point of each stage versus the logarithm of the cut-point of each stage.

TABLE 2

Particle size distribution - individual and sample stage values

Group	Week	% of total collected on each stage							Particle size		
		3	4	5	6	7	8	Filter	MMAD	σg	% < 7μm
5	1	0.0	17.2	38.6	38.6	4.1	1.4	0.0	3.1	1.60	96
(High dose	2	2.5	15.8	50.8	23.3	5.8	0.8	0.8	3.5	1.95	85
Cryolite)	3	0.0	23.7	42.4	18.6	7.6	1.7	5.9	2.5	2.03	93
	4	4.3	34.3	31.9	12.6	4.3	3.9	8.7	3.3	2.90	76
	5	0.0	12.7	45.8	34.5	3.5	0.7	2.8	2.6	1.83	95
	6	1.4	27.8	35.4	22.5	3.3	1.0	8.6	2.9	2.42	84
	7	0.6	12.9	35.5	34.8	3.9	1.9	10.3	2.3	2.23	92
	8	1.1	14.4	35.0	33.3	5.6	3.3	7.2	2.5	2.24	90
	9	2.8	13.6	37.5	33.0	6.3	2.8	4.0	2.8	2.20	87
	10	5.2	14.0	30.1	31.0	7.4	4.8	7.4	2.7	2.58	84
	11	0.0	16.1	37.1	31.7	4.8	5.4	4.8	2.3	1.97	95
	12	6.9	35.6	17.2	23.0	10.3	3.4	3.4	3.7	2.61	75
	13	1.3	13.9	45.6	13.9	7.6	11.4	6.3	2.4	2.35	90
	Mean	2.0	19.4	37.1	27.0	5.7	3.3	5.4	2.8	2.30	86

MMAD Mass median aerodynamic diameter
σg standard deviation

Collection characteristics

Marple personal cascade impactor cut-points at 2 l/minute

Impactor stage	3.00	4.00	5.00	6.00	7.00	8.00	Filter
Cut-point* (μm)	9.80	6.00	3.50	1.55	0.93	0.52	0.00

* Aerodynamic equivalent particle for spherical particles of unit mass density in air at 25°C and 760mmHg

Calculation of MMAD, σg and inhalable fraction (% < 7μm)

By linear regression - probit of cumulative percentage , by mass , of particles smaller than cut-point of each stage versus the logarithm of the cut-point of each stage.

TABLE 3

Estimates of nominal concentration

Group	Exposure	Canister Initial weight (g)	Canister Final weight (g)	Total (g)	Time (mins)	Nominal concentration (mg/m^3)
2 (NaF)	1 - 7[1]	357.24	346.62	10.62	2520	(70)[1]
	8 - 16[2]	355.95	349.07	6.88	2940	39
	16 - 22[3]	356.18	351.26	4.92	2130	38
	22 - 25	355.60	352.52	3.08	1410	36
	26	356.53	355.59	0.94	360	44
	27 - 33	356.06	348.74	7.32	2520	48
	34 - 38	355.59	350.37	5.22	1800	48
	39 - 44	355.46	349.23	6.23	2160	48
	45 - 49	356.34	351.22	5.12	1800	47
	50 - 53[4]	355.35	352.23	3.12	1260	46
	53[4] - 58	356.93	350.62	6.31	1980	53
	59 - 64	355.59	349.80	5.79	2040	47
	65 - 66	356.55	353.77	2.78	720	64
					Mean	47
					sd	7.39
3 (Low dose Cryolite)	1 - 66	357.55	356.35	1.2	23640	0.85
4 (Inter dose Cryolite)	1 - 48[5]	357.77	353.00	4.77	17280	4.6
	48 - 66	353.67	352.07	1.6	6480	4.4
					Average	4.5
5 (High dose Cryolite)	1 - 12	358.45	351.70	6.75	4320	26
	13[1]	353.66	352.70	0.96	90	(178)[1]
	13 - 15	351.70	350.50	1.20	990	20
	16 - 30	354.37	347.20	7.17	5460	22
	31 - 42	358.41	352.21	6.20	4320	24
	43	353.58	352.56	1.02	360	(47)[1]
	44 - 57	358.89	351.69	7.20	5040	24
	58 - 66	357.23	352.65	4.58	3120	24
					Mean	23.3
					sd	2.07

sd Standard deviation

1 Gross fragmentation of the dust cake resulting in loss of test material, weights not used in calculation of the study mean

2 1 hour into the exposure a blockage in the WDFM occurred and the WDF canister was changed

3 0.5 hour into the exposure a blockage in the WDFM occurred and the WDF canister was changed

4 During the exposure the airflow to WDFM dropped indicating a possible blockage. The WDF canister was changed

5 2 hour into the exposure the WDFM was not functioning and was changed

Nominal concentration was calculated as follows:

$$\text{Total particulate } (mg/m^3) = \frac{w \times 1000}{t \times f/1000}$$

Where w = powder usage (g)
t = duration of exposure
f = chamber airflow (60 l/minute)

TABLE 4

Exposure mean chamber temperature (°C)

Exposure	Group				
	1 Air control	2 NaF	3 Low dose Cryolite	4 Inter dose Cryolite	5 High dose Cryolite
1	23.7	22.8	21.2	21.4	22.8
2	23.7	22.8	21.2	21.7	22.8
3	22.8	22.3	20.9	21.0	22.5
4	23.7	22.8	20.8	21.5	22.8
5	23.7	22.8	20.8	20.9	22.7
6	23.6	22.8	20.9	21.5	22.7
7	23.3	22.8	20.9	21.3	22.6
8	23.4	23.1	21.2	21.5	22.8
9	24.0	23.6	21.8	22.0	23.3
10	23.3	22.7	20.9	21.4	22.2
11	23.2	23.0	21.2	21.6	22.6
12	22.8	22.6	20.8	20.9	22.1
13	23.5	23.0	21.3	21.5	22.3
14	23.4	22.8	21.4	21.4	22.3
15	22.8	22.7	20.8	20.9	22.1
16	23.0	22.7	21.0	21.3	21.8
17	23.0	22.7	20.9	21.2	22.5
18	22.8	22.5	20.8	21.2	22.1
19	22.8	22.5	20.8	21.2	22.0
20	23.1	22.5	20.8	21.2	22.2
21	23.1	22.9	21.1	21.2	22.4
22	23.3	22.8	20.9	21.3	22.3
23	23.3	22.9	20.9	21.4	22.2
24	23.2	22.7	20.9	21.7	22.7
25	23.5	22.8	21.0	21.5	22.2
26	23.5	22.7	21.1	21.3	22.3
27	23.2	22.7	21.0	21.6	22.3
28	23.8	23.7	21.7	22.2	23.0
29	23.7	23.3	21.3	22.2	22.7
30	23.3	23.1	20.9	21.7	22.5
31	23.7	23.2	20.9	21.6	22.3
32	23.2	23.1	21.2	21.4	22.5
33	23.6	23.3	21.3	21.4	22.2

TABLE 4

(Exposure mean chamber temperature - continued)

Exposure	Group				
	1 Air control	2 NaF	3 Low dose Cryolite	4 Inter dose Cryolite	5 High dose Cryolite
34	23.3	23.1	20.9	21.7	22.5
35	23.5	23.2	21.2	21.7	22.5
36	23.6	23.1	21.5	21.7	22.7
37	23.5	23.1	21.3	21.7	22.5
38	23.7	23.0	20.9	21.5	22.2
39	23.6	22.9	20.9	21.6	22.1
40	23.5	23.0	21.2	21.7	22.2
41	23.7	22.8	21.0	21.6	22.5
42	23.3	22.8	21.0	21.5	22.4
43	24.5	23.1	21.2	21.6	22.5
44	23.6	23.3	21.1	21.7	22.3
45	23.8	22.8	20.9	21.8	22.3
46	23.7	23.0	20.9	21.5	22.3
47	23.6	23.0	21.0	21.6	22.2
48	24.0	23.5	21.5	22.0	22.8
49	23.7	23.5	21.2	21.8	22.8
50	23.7	23.1	21.1	21.7	22.0
51	23.6	22.7	21.2	21.6	22.2
52	23.7	22.8	21.2	21.3	22.6
53	23.6	23.2	20.9	21.7	22.7
54	23.3	23.0	21.0	21.3	22.3
55	23.5	23.1	21.3	21.7	22.2
56	23.7	23.0	21.1	21.7	22.7
57	23.7	22.8	21.8	21.3	22.2
58	23.7	22.6	20.8	21.3	22.3
59	23.3	22.8	20.9	21.0	22.3
60	23.7	22.8	21.3	21.5	22.2
61	23.4	22.8	20.9	21.8	22.3
62	23.3	22.4	20.5	21.0	21.9
63	23.5	23.1	21.3	21.1	21.7
64	23.4	22.8	20.9	21.4	21.7
65	23.5	23.2	21.3	21.4	22.3
66	20.8	20.2	20.0	20.2	19.8
Mean	23.4	22.9	21.1	21.5	22.3
sd	0.45	0.43	0.30	0.32	0.44

sd Standard deviation

TABLE 5

Bodyweights - group mean values (g)

Week	Group									
	1M Air Control	2M NaF	3M Low dose	4M Inter dose	5M High dose	1F Air Control	2F NaF	3F Low dose	4F Inter dose	5F High dose
-1	205	204	206	208	204	178	174	178	177	175
0	260	260	260	265	257	201	199	197	197	197
1	283	284	284	288	278	211	206	203	205	205
2	308	311	306	309	301	220	217	212	210	215
3	329	336	331	331	320	230	225	222	220	225
4	347	356	352	352	338	236	234	228	229	232
5	365	372	370	371	356	245	239	237	234	237
6	380	390	392	391	373	251	247	246	240	242
7	396	404	405	407	386	256	252	252	245	248
8	411	419	417	417	399	258	257	253	252	251
9	425	431	430	432	407	263	259	259	250	254
10	435	444	435	441	418	266	262	259	255	253
11	445	452	445	448	427	270	264	264	260	258
12	449	459	445	454	429	270	265	266	259	261
13	445	456	443	448	427	268	262	259	256	256
13	456	465			425	271	262			259
14	475	485			451	281	276			279
15	496	506			469	289	282			290
16	511	520			485	293	290			299
17	523	532			496	298	293			302
18	534	544			505	303	297			310
19	542	556			515	306	297			311
20	552	565			522	308	303			317
21	562	573			532	313	305			318
22	573	584			538	315	309			321
23	585	595			550	318	312			325
24	590	601			555	326	315			328
25	599	612			565	327	317			327
26	598	613			562	326	317			324
Gain										
1 to 13	185	196	183	183	170	67	62	63	59	59
13 to 26	143	148			137	55	55			59

No differences of statistical significance

TABLE 6

Food consumption - group mean values (g)

Week	Group									
	1M Air Control	2M NaF	3M Low dose	4M Inter dose	5M High dose	1F Air Control	2F NaF	3F Low dose	4F Inter dose	5F High dose
-1	212	209	209	220	207	158	156	151	156	152
1	197	198	194	201	186	150	143	143	147	144
2	187	196	196	192	189	151	146	144	149	149
3	195	202	202	195	190	153	147	146	154	150
4	195	210	206	204	196	158	150	149	159	153
5	198	207	209	204	198	158	151	155	158	154
6	204	211	218	212	203	159	154	159	164	157
7	203	212	214	215	205	158	151	159	158	157
8	203	207	213	204	200	154	148	156	157	153
9	205	211	210	211	198	156	148	152	155	153
10	202	209	209	206	199	154	148	147	153	151
11	200	208	205	201	197	152	145	148	154	151
12	200	211	204	205	195	150	148	150	151	150
13	174	187	179	180	177	136	133	128	131	133
14	217	220			204	159	160			169
15	224	226			213	161	160			175
16	218	218			211	160	156			171
17	218	221			210	161	153			169
18	219	221			207	161	153			169
19	216	224			209	160	150			165
20	218	220			210	160	154			166
21	216	217			208	159	152			163
22	218	216			206	157	150			156
23	225	227			217	165	159			168
24	218	223			211	170	154			159
25	216	221			211	162	155			159
26	195	199			189	151	144			147
Cumulative										
1 to 13	2562	2668	2656	2629	2533	1990	1914	1935	1991	1954
14 to 26	2817	2854			2706	2085	2000			2132

No differences of statistical significance

TABLE 7

Water consumption - group mean values (g)

Week	Group									
	1M Air Control	2M NaF	3M Low dose	4M Inter dose	5M High dose	1F Air Control	2F NaF	3F Low dose	4F Inter dose	5F High dose
-1	216.7	217.8	211.9	228.4	217.9	167.3	162.7	164.7	182.7	167.8
1	214.2	216.5	221.7	225.3	209.4	173.3	165.8	168.0	171.7	166.0
2	203.8	215.3	212.4	200.6	202.5	169.2	170.8	169.8	165.4	163.5
3	206.9	216.1	219.5	200.1	203.2	170.4	172.3	189.5	166.3	162.7
4	209.8	224.6	226.8	203.9	211.0	171.9	174.9	198.6	171.6	182.5
5	214.1	230.8	225.6	207.3	217.4	177.5	177.9	197.7	174.5	192.2
6	221.6	229.7	233.8	212.0	226.5	178.6	180.0	214.0	176.6	194.2
7	229.7	243.1	239.2	221.3	232.2	181.7	184.8	208.1	179.5	170.0
8	221.3	234.6	232.8	215.2	231.2	173.3	180.3	184.5	174.8	189.5
9	229.1	242.4	240.3	226.0	231.5	182.4	187.8	202.2	180.7	207.9
10	234.8	246.8	242.9	226.2	236.9	186.9	190.7	193.5	188.9	225.3
11	239.3	250.8	245.1	228.1	239.8	197.1	193.4	208.6	190.2	246.2
12	238.4	247.0	257.0	231.8	232.0	191.9	201.3	201.6	186.0	254.5
13	230.4	249.2	227.0	211.7	234.0	190.4	185.9	170.7	170.4	205.7
14	239.5	251.3			239.9	187.0	201.9			201.1
15	241.3	249.4			240.9	193.5	215.1			217.8
16	237.1	244.3			245.4	202.1	209.8			250.1
17	224.2	243.8			236.3	201.9	209.1			223.2
18	231.4	245.3			231.6	204.2	208.7			237.4
19	234.7	244.1			233.9	202.2	205.7			232.1
20	244.0	235.6			238.8	206.4	212.8			224.8
21	241.1	241.2			234.5	208.2	213.3			237.7
22	227.6	228.8			227.0	227.6	208.9			203.4
23	225.3	223.5			217.8	223.5	202.2			199.2
24	223.0	224.7			215.8	219.3	207.9			186.1
25	228.6	229.6			213.9	226.3	213.8			191.2
26	197.6	210.3			202.9	205.4	189.3			190.2
Cumulative										
1 to 13	2893	3047	3024	2810	2907	2344	2361	2507	2297	2560
14 to 26	2995	3072			2979	2708	2699			2734

TABLE 8

Haematology - group mean values

Week 13

Group	PCV %	Hb g/dl	RBC $10^{12}/l$	MCHC g/dl	MCV fl	MCH pg	Retic %	WBC Total $10^9/l$	N $10^9/l$	L $10^9/l$	E $10^9/l$	B $10^9/l$	M $10^9/l$	LUC $10^9/l$	Plt $10^9/l$
1M Air Control	44.9	15.8	8.62	35.3	52.1	18.4	1.4	10.97	1.91	8.61	LT 0.23	LT 0.03	NP 0.11 (0.08)	0.10	972
2M NaF	44.1	15.4	8.42	34.9	52.4	18.3	+ 2.1	11.79	1.87	9.23	0.17	0.03	0.39 (0.12)	0.09	969
3M Low dose	45.4	15.9	8.68	35.0	52.3	18.3	1.6	9.29	1.75	7.26	0.13	0.02	0.08 (0.07)	0.06	931
4M Inter dose	45.6	15.8	8.60	34.7	53.0	18.4	** 2.4	12.55	2.66	9.42	0.24	0.03	0.12 (0.12)	0.08	962
5M High dose	45.1	15.8	8.80	35.0	51.3	18.0	** 2.2	* 14.46	2.19	* 11.66	0.30	* 0.05	0.15 (0.13)	0.11	903

LT Log-transformed data used in the analysis
NP Non-parametric analysis employed. Median values shown in parentheses
Level of significance - all comparisons made with Air control
Williams test: * $p<0.05$
** $p<0.01$
Students 't' test: + $p<0.05$

TABLE 8

(Haematology - continued)

Week 13

Group	PCV	Hb	RBC	MCHC	MCV	MCH	Retic	WBC Total	N	L	E	B	M	LUC	Plt
	%	g/dl	10^{12}/l	g/dl	fl	pg	%	10^9/l	10^9/l	10^9/l	10^9/l	10^9/l	10^9/l	10^9/l	10^9/l
1F Air Control	42.1	15.1	7.64	35.9	55.2	19.8	1.3	LT 7.91	0.99	LT 6.64	0.14	LT 0.02	0.07	0.05	1043
2F NaF	42.9	15.2	7.89	+ 35.5	54.3	19.3	1.6	9.06	0.84	7.96	0.12	0.02	0.07	0.05	971
3F Low dose	42.6	15.0	7.66	** 35.3	55.7	19.6	2.0	7.12	1.09	5.70	0.16	0.01	0.08	0.09	994
4F Inter dose	43.0	15.2	7.85	** 35.3	54.9	19.3	1.9	* 10.33	* 1.38	8.57	0.19	0.02	0.09	0.08	936
5F High dose	43.2	15.2	7.84	** 35.2	55.1	19.4	1.6	** 12.04	** 1.55	* 10.01	0.19	** 0.04	** 0.13	** 0.13	934

LT Log-transformed data used in the analysis

Level of significance - all comparisons made with Air control

Williams test: * $p<0.05$

** $p<0.01$

Students 't' test: + $p<0.05$

TABLE 9

Biochemistry - group mean values

Week 13

Group	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	GOT mU/ml
1M Air Control	106	6.8	2.9	3.9	16	0.6	179	32	68
2M NaF	109	6.7	+ 2.8	4.0	15	0.6	186	32	75
3M Low dose	117	6.7	2.8	3.9	17	0.6	183	31	72
4M Inter dose	* 122	6.8	2.9	3.9	16	0.5	176	28	65
5M High dose	* 118	6.8	2.9	3.9	16	0.5	190	30	69

Level of significance - all comparisons made with Air control
Williams test: * $p<0.05$
Students 't' test: + $p<0.05$

TABLE 9

(Biochemistry - continued)

Week 13

Group	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	GOT mU/ml
1F Air Control	112	7.2	3.2	4.0	21	0.7	LT 106	LT 27	LT 66
2F NaF	122	7.3	3.3	4.0	19	0.6	89	28	66
3F Low dose	118	7.3	3.3	4.0	21	0.7	100	34	77
4F Inter dose	114	7.2	3.3	3.9	19	0.6	102	39	79
5F High dose	125	7.4	3.4	4.1	18	0.6	92	39	79

LT Log transformed data used in the analysis
No differences of statistical significance

TABLE 9

(Biochemistry - continued)

Week 13

Group	γGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
1M Air Control	FA <1	109	0.2	146	3.4	5.4	3.7	103	62
2M NaF	<1	98	0.1	146	3.5	5.4	3.7	104	55
3M Low dose	<1	102	0.2	147	* 3.6	5.4	3.8	104	55
4M Inter dose	<1	117	0.1	147	** 3.8	5.5	* 4.0	104	62
5M High dose	<1	107	<0.2	147	** 3.7	5.5	** 4.2	104	62

FA Frequency analysis applied to the data
Level of significance - all comparisons made with Air control
Williams test: * $p<0.05$
** $p<0.01$

TABLE 9

(Biochemistry - continued)

Week 13

Group	γGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
1F Air Control	<1	LT 80	0.2	145	3.1	5.4	3.1	103	75
2F NaF	<1	81	0.2	145	+ 3.5	5.5	3.5	104	70
3F Low dose	<1	202	0.2	146	* 3.5	5.5	3.0	104	72
4F Inter dose	<1	165	0.2	145	** 3.5	5.5	3.5	103	73
5F High dose	<1	88	0.2	** 147	** 3.6	** 5.6	** 3.9	* 105	87

LT Log transformed data used in the analysis

Level of significance - all comparisons made with Air control

Williams test: * $p<0.05$

** $p<0.01$

Students 't' test: + $p<0.05$

TABLE 10

Urinalysis - group mean values

Week 13

Group	Volume ml	pH	SG	Protein mg/dl
1M Air Control	4.7	6.8	1046	219
2M NaF	4.8	7.1	1045	211
3M Low dose	5.1	* 7.3	1040	205
4M Inter dose	6.8	* 7.2	1035	163
5M High dose	4.3	* 7.2	1042	250

Level of sigificance - all comparisons made with Air control
Williams test: * $p<0.05$

TABLE 10

(Urinalysis - continued)

Week 13

Group	Volume ml	pH	SG	Protein mg/dl
1F Air Control	3.2	6.4	1046	88
2F NaF	3.6	6.4	1043	78
3F Low dose	3.2	6.4	1044	78
4F Inter dose	4.0	6.5	1042	82
5F High dose	3.8	6.4	1044	81

No differences of statistical significance

TABLE 11

Urinary fluoride and aluminium - group mean values

Male rats

Group	Concentration (mg/litre)			
	Fluoride		Aluminium	
	Terminal	Withdrawal	Terminal	Withdrawal
1 (Air control)	1.5	1.05	1.5	<0.5
2 (NaF)	6.25	1.5	0.5	<0.5
3 (Low dose Cryolite)	1.25	-	<0.5	-
4 (Inter dose Cryolite)	1.4	-	0.6	-
5 (High dose Cryolite)	3.8	0.95	1.8	<0.5

Female rats

Group	Concentration (mg/litre)			
	Fluoride		Aluminium	
	Terminal	Withdrawal	Terminal	Withdrawal
1 (Air control)	1.35	1.08	<0.6	<0.5
2 (NaF)	7.25	1.3	<0.6	0.24
3 (Low dose Cryolite)	1.4	-	2.5	-
4 (Inter dose Cryolite)	1.5	-	1.1	-
5 (High dose Cryolite)	4.6	1.2	2.6	<0.5

TABLE 12

Bone fluoride and aluminium - group mean values

Male rats

Group	Concentration (%w/w)			
	Fluoride		Aluminium	
	Terminal	Withdrawal	Terminal	Withdrawal
1 (Air control)	0.026	0.027	<0.01	<0.01
2 (NaF)	0.057	0.054	<0.01	<0.01
3 (Low dose Cryolite)	0.022	-	<0.01	-
4 (Inter dose Cryolite)	0.018	-	<0.01	-
5 (High dose Cryolite)	0.041	0.039	<0.01	<0.01

Female rats

Group	Concentration (%w/w)			
	Fluoride		Aluminium	
	Terminal	Withdrawal	Terminal	Withdrawal
1 (Air control)	0.032	0.037	<0.01	<0.01
2 (NaF)	0.079	0.073	<0.01	<0.01
3 (Low dose Cryolite)	0.030	-	<0.01	-
4 (Inter dose Cryolite)	0.028	-	<0.01	-
5 (High dose Cryolite)	0.063	0.064	<0.01	<0.01

TABLE 13

Tooth fluoride and aluminium - group mean values

Male rats

Group	Concentration (%w/w)			
	Fluoride		Aluminium	
	Terminal	Withdrawal	Terminal	Withdrawal
1 (Air control)	0.015	0.013	<0.01	<0.01
2 (NaF)	0.020	0.011	<0.01	<0.01
3 (Low dose Cryolite)	0.009	-	<0.01	-
4 (Inter dose Cryolite)	0.017	-	<0.01	-
5 (High dose Cryolite)	0.017	0.010	<0.01	<0.01

Female rats

Group	Concentration (%w/w)			
	Fluoride		Aluminium	
	Terminal	Withdrawal	Terminal	Withdrawal
1 (Air control)	0.018	0.016	<0.01	<0.01
2 (NaF)	0.018	0.013	<0.01	<0.01
3 (Low dose Cryolite)	0.013	-	<0.01	-
4 (Inter dose Cryolite)	0.018	-	<0.01	-
5 (High dose Cryolite)	0.023	0.013	<0.01	<0.01

TABLE 14

Macroscopic pathology incidence summary - termination

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Fur										
Coarse - dorsum	0	0	0	0	0	0	0	1	0	0
Skin										
Scab/s	0	1	0	0	0	0	0	0	0	0
Skin										
Alopecia	0	3	0	0	0	1	2	3	1	0
Tail										
Malaligned	0	1	0	0	0	0	0	0	0	0
Lymph Nodes - Cervical										
Congested	0	2	0	0	0	0	0	1	0	0
Lungs										
Congested	3	1	0	0	0	0	0	0	0	0
Pale subpleural foci	1	1	0	0	0	2	0	1	1	0
Adipose Tissue										
Minimal	0	0	0	0	0	0	0	1	0	0
Liver										
Median cleft, pale subcapsular area/s	2	1	1	0	0	0	0	0	1	0
Pale subcapsular area/s	0	0	0	0	1	0	0	0	0	0
Congested lobe/s	0	0	0	0	0	0	0	0	1	0
Firm lobe/s	0	0	0	0	0	0	0	0	1	0

TABLE 14

(Macroscopic pathology incidence summary - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Forestomach										
Cyst - limiting ridge	0	0	2	0	0	1	0	1	0	0
Stomach Corpus Mucosa										
White nodule	0	0	0	0	1	0	0	0	0	0
Stomach Antrum Mucosa										
White nodule/s	0	2	0	0	0	1	1	1	0	1
Kidneys										
Increased pelvic dilatation	0	2	0	0	0	0	0	0	0	1
Epididymides										
Yellow swelling/s	0	0	0	1	0	0	0	0	0	0
Uterus										
Fluid distension	0	0	0	0	0	3	3	1	1	2

TABLE 15

Macroscopic pathology incidence summary - withdrawal

Removal reason: Recovery	Group 1	Group 2	Group 5	Group 1	Group 2	Group 5
	-------- Males --------			------- Females -------		
Animals on study	20	20	20	20	20	20
Animals completed	10	10	10	10	10	9
Skin						
Scab/s	1	1	2	1	0	0
Skin						
Alopecia	4	1	2	1	2	5
Subcutis						
Mass	0	0	0	1	0	1
Tail						
Pustule/s	0	0	0	1	0	0
Pituitary						
Swollen	0	0	0	1	1	0
Lymph Nodes - Cervical						
Congested	1	0	0	0	0	0
Lymph Nodes - Mediastinal						
Enlarged	1	0	0	0	0	0
Congested	0	0	0	1	0	0
Lungs						
Congested	3	4	3	0	3	3
Pale subpleural foci	2	4	3	1	4	2
Lymph Nodes - Tracheobronchial						
Enlarged	6	4	5	1	1	2

TABLE 15

(Macroscopic pathology incidence summary - continued)

Removal reason: Recovery	Group 1	Group 2	Group 5	Group 1	Group 2	Group 5
	-------- Males --------			------- Females -------		
Animals on study	20	20	20	20	20	20
Animals completed	10	10	10	10	10	9
Liver						
Median cleft, pale subcapsular area/s	1	2	0	1	0	2
Forestomach						
Cyst - limiting ridge	0	0	1	0	0	0
A caseous cyst	1	0	0	0	0	0
Stomach Antrum Mucosa						
White nodule/s	0	2	1	1	0	0
Kidneys						
Increased pelvic dilatation	0	2	0	0	1	1
Irregular cortical scarring	0	0	0	0	1	0
Pelvis contained cloudy fluid	0	1	0	0	0	0
Ureters						
Distended	0	0	0	0	1	0
Epididymides						
Pale area	0	0	1	0	0	0
Ovaries						
No corpora lutea visible	0	0	0	0	0	1
Uterus						
Fluid distension	0	0	0	2	3	1

TABLE 16

Organ weights - group mean values - termination

Week 14

Group	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg	Testes L g	Testes R g	Epididymides Left g	Epididymides Right g
1M		A	A/LT	A	A	A	A	A	A
Air	425	1.46	13.3	2.38	51.8	1.72	1.69	0.593	0.581
Control		(1.48)	(13.6)	(2.43)	(52.6)	(1.73)	(1.70)	(0.590)	(0.590)
2M									
NaF	441	1.48	13.4	2.55	61.5	1.77	1.73	0.617	0.620
		(1.46)	(13.0)	(2.52)	(60.9)	(1.76)	(1.72)	(0.613)	(0.614)
3M									
Low	437	1.47	14.0	2.64	53.8	1.69	1.60	0.581	0.564
dose		(1.46)	(13.8)	(2.63)	(53.6)	(1.69)	(1.60)	(0.580)	(0.561)
4M									
Inter	444	1.52	13.8	2.58	53.9	1.64	1.58	0.588	0.548
dose		(1.50)	(13.4)	(2.53)	(53.0)	(1.63)	(1.56)	(0.582)	(0.539)
5M		**							
High	425	1.68	13.9	2.53	54.2	1.69	1.64	0.547	0.580
dose		(1.70)	(14.2)	(2.58)	(55.1)	(1.70)	(1.66)	(0.553)	(0.599)

A Bodyweight used as a covariate. Adjusted values shown in parentheses
LT Log-transformed data used in the analysis
Level of signficance - all comparisons made with Air control
Williams' test ** $p<0.01$

TABLE 16

(Organ weights - group mean values - continued)

Week 14

Group	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg
1F Air Control	261	A 1.12 (1.10)	A 8.9 (8.7)	A 1.70 (1.67)	A 62.6 (61.8)
2F NaF	257	1.15 (1.14)	8.6 (8.6)	1.69 (1.68)	65.0 (64.6)
3F Low dose	250	1.17 (1.18)	8.8 (8.9)	1.68 (1.70)	70.5 (71.1)
4F Inter dose	252	1.18 (1.19)	8.7 (8.8)	1.73 (1.74)	71.2 (71.5)
5F High dose	254	** 1.27 (1.27)	8.7 (8.7)	1.57 (1.58)	66.2 (66.3)

A Bodyweight used as a covariate. Adjusted values shown in parentheses
Level of signifcance: all comparisons made with Air control
Williams' test: ** $p<0.01$

TABLE 17

Organ weights - group mean values - withdrawal

Week 27

Group	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg	Testes L g	Testes R g	Epididymides Left g	Epididymides Right g
1M Air Control	594	A 1.80 (1.78)	A 17.4 (17.2)	A 3.07 (3.03)	LT/A 51.1 (49.9)	1.78	1.79	0.652	0.705
2M NaF	608	2.04 (2.00)	19.1 (18.4)	3.24 (3.13)	56.3 (52.8)	1.83	1.82	0.699	0.716
5M High dose	557	++ 2.07 (2.13)	17.4 (18.3)	++ 3.21 (3.36)	53.0 (55.6)	1.85	1.86	0.693	0.688

A Bodyweight used as a covariate in the analysis. Median values shown in parentheses
LT Log-transformed data used in the analysis
Level of significance - all comparisons made with Air control
Student's 't' test: ++ $p<0.01$

TABLE 17

Organ weights - group mean values

Week 27

Group	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg
1F Air Control	323	A 1.40 (1.39)	A 11.7 (11.6)	A 2.08 (2.06)	75.9
2F NaF	310	1.47 (1.48)	11.6 (11.9)	1.96 (1.99)	67.3
5F High dose	321	1.55 (1.55)	11.3 (11.2)	2.08 (2.07)	69.3

A Bodyweight used as a covariate in the analysis. Adjusted values shown in parentheses

No differences of statistical significance

TABLE 18

Microscopic pathology incidence summary - intercurrent

Removal reason: Intercurrent	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	0	0	0	0	0	1	1	0	0	0
Nasal Passages										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Pharynx										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Larynx										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	0	0	0	0
Epithelial hyperplasia - ventral (Total)	0	0	0	0	0	0	1	0	0	0
Moderate	0	0	0	0	0	0	1	0	0	0
Trachea (including Bifurcation)										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Lungs										
Examined	0	0	0	0	0	1	1	0	0	0
Vascular congestion (Total)	0	0	0	0	0	1	1	0	0	0
Minimal	0	0	0	0	0	1	1	0	0	0
Aorta										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Heart										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Thymus										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0

TABLE 18

(Microscopic pathology incidence summary - intercurrent - continued)

Removal reason: Intercurrent	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	0	0	0	0	0	1	1	0	0	0
Lymph Nodes - Cervical										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Lymph Nodes - Mesenteric										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Lymph Nodes - Tracheobronchial										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Lymph Nodes - Mediastinal										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Spleen										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Liver										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Pancreas										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Kidneys										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	0	1	0	0	0
Dystrophic mineralisation (Total)	0	0	0	0	0	1	0	0	0	0
Trace	0	0	0	0	0	1	0	0	0	0
Urinary Bladder										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0

TABLE 18

(Microscopic pathology incidence summary - intercurrent - continued)

Removal reason: Intercurrent	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	0	0	0	0	0	1	1	0	0	0
Uterus										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Cervix										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Ovaries										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Thyroids										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Parathyroids										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Adrenals										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Pituitary										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Salivary Glands										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Oesophagus										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0

TABLE 18

(Microscopic pathology incidence summary - intercurrent - continued)

Removal reason: Intercurrent	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	0	0	0	0	0	1	1	0	0	0
Stomach										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Duodenum										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Jejunum										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Ileum										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Caecum										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Colon										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Rectum										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Eyes										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Sciatic Nerve										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0

TABLE 18

(Microscopic pathology incidence summary - intercurrent - continued)

Removal reason: Intercurrent	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	0	0	0	0	0	1	1	0	0	0
Brain										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Sternum										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	1	1	0	0	0
Factors Contributory To Death										
Examined	0	0	0	0	0	1	1	0	0	0
Unknown	0	0	0	0	0	1	1	0	0	0

TABLE 19

Microscopic pathology incidence summary - termination

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Nasal Passages										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	9	9	0	0	10	9	9	0	0	10
Degeneration of olfactory epithelium (Total)	1	1	0	0	0	0	0	0	0	0
Trace	0	1	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Pharynx										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Larynx										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	9	0	0	0	10	9	1	0	0	10
Epithelial hyperplasia - arytenoids (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Necrosis of ventral cartilage	0	1	0	0	0	0	0	0	0	0
Epithelial hyperplasia - ventral (Total)	0	8	0	0	0	0	8	0	0	0
Trace	0	2	0	0	0	0	2	0	0	0
Minimal	0	3	0	0	0	0	5	0	0	0
Moderate	0	3	0	0	0	0	1	0	0	0
Subepithelial inflammation - ventral (Total)	0	5	0	0	0	0	4	0	0	0
Trace	0	2	0	0	0	0	2	0	0	0
Minimal	0	2	0	0	0	0	2	0	0	0
Moderate	0	1	0	0	0	0	0	0	0	0
Epithelial mineralisation - ventral (Total)	0	1	0	0	0	0	0	0	0	0
Moderate	0	1	0	0	0	0	0	0	0	0
Trachea (including Bifurcation)										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Lungs										
Examined	10	10	10	10	10	9	9	10	10	10
No abnormalities detected	5	3	7	5	0	6	6	10	5	0
Pneumonitis (Total)	0	2	0	0	0	0	0	0	0	0
Trace	0	2	0	0	0	0	0	0	0	0
Perivascular inflammatory infiltration(Total)	1	0	0	0	2	1	0	0	0	0
Trace	0	0	0	0	1	1	0	0	0	0
Minimal	1	0	0	0	1	0	0	0	0	0
Subpleural aggregation of alveolar macrophages(Total)	1	1	2	0	0	2	1	0	0	0
Trace	0	1	1	0	0	2	0	0	0	0
Minimal	1	0	1	0	0	0	1	0	0	0
Vascular congestion (Total)	1	0	0	0	0	0	0	0	1	0
Trace	1	0	0	0	0	0	0	0	0	0
Minimal	0	0	0	0	0	0	0	0	1	0
Aggregates of macrophages (Total)	1	4	2	0	0	0	1	0	0	0
Trace	1	2	2	0	0	0	1	0	0	0
Minimal	0	2	0	0	0	0	0	0	0	0
Recent alveolar haemorrhage (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Macrophages around alveolar ducts (Total)	0	0	0	0	9	0	0	0	0	6
Trace	0	0	0	0	2	0	0	0	0	4
Minimal	0	0	0	0	5	0	0	0	0	2
Moderate	0	0	0	0	2	0	0	0	0	0
Eosinophilic crystals assoc macrophages around alveolar ducts (Total)	0	1	0	0	0	0	0	0	0	0
Minimal	0	1	0	0	0	0	0	0	0	0
Increased collagen in alveolar duct walls(Total)	0	0	0	0	2	0	0	0	0	0
Trace	0	0	0	0	2	0	0	0	0	0

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Lungs	(Continued)									
Extension of bronchiolar epithelium into alveolar ducts(Total)	0	0	0	0	3	0	0	0	0	0
Trace	0	0	0	0	2	0	0	0	0	0
Minimal	0	0	0	0	1	0	0	0	0	0
Alveolitis with interstitial thickening of alveolar duct walls (Total)	0	0	0	5	10	0	1	0	3	8
Trace	0	0	0	5	5	0	1	0	3	6
Minimal	0	0	0	0	5	0	0	0	0	2
Mural hypertrophy of bronchial arteries	0	0	0	0	1	0	0	0	0	0
Brown pigment in macrophages (Total)	0	0	0	1	9	0	0	0	1	8
Trace	0	0	0	1	7	0	0	0	1	7
Minimal	0	0	0	0	2	0	0	0	0	1
Macrophages containing fine brown pigment in BALT(Total)	0	0	0	0	5	0	0	0	0	1
Trace	0	0	0	0	5	0	0	0	0	1
Ectopic bone focus	0	0	0	0	0	0	0	0	1	0
Aorta										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Heart										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	5	4	0	0	4	7	9	0	0	9
Myocarditis (Total)	4	5	0	0	6	1	0	0	0	1
Trace	3	4	0	0	5	1	0	0	0	1
Minimal	1	1	0	0	1	0	0	0	0	0
Myocardial fibrosis (Total)	0	0	0	0	0	1	0	0	0	0
Minimal	0	0	0	0	0	1	0	0	0	0

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Heart	(Continued)									
Myocardial necrosis (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Valvular cyst	0	1	0	0	0	0	0	0	0	0
Valvular endocarditis (Total)	0	0	0	0	1	0	0	0	0	0
Minimal	0	0	0	0	1	0	0	0	0	0
Thymus										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	8	10	0	0	10	9	9	0	0	10
Involution/atrophy (Total)	2	0	0	0	0	0	0	0	0	0
Trace	2	0	0	0	0	0	0	0	0	0
Lymph Nodes - Cervical										
Examined	10	10	0	0	10	9	9	1	0	10
No abnormalities detected	10	7	0	0	9	9	9	0	0	10
Increased cellularity - generalised (Total)	0	1	0	0	0	0	0	0	0	0
Minimal	0	1	0	0	0	0	0	0	0	0
Sinus erythrocytosis (Total)	0	0	0	0	1	0	0	1	0	0
Minimal	0	0	0	0	1	0	0	1	0	0
Sinus histiocytosis (Total)	0	2	0	0	0	0	0	0	0	0
Minimal	0	2	0	0	0	0	0	0	0	0
Lymph Nodes - Mesenteric										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Lymph Nodes - Tracheobronchial										
Examined	10	9	9	9	9	8	7	9	8	10
Missing	0	1	1	1	1	1	2	1	2	0
No abnormalities detected	10	9	9	8	2	8	6	9	8	4

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Lymph Nodes - Tracheobronchial	(Continued)									
Macrophages containing brown pigment (Total)	0	0	0	1	7	0	0	0	0	6
Trace	0	0	0	1	1	0	0	0	0	4
Minimal	0	0	0	0	5	0	0	0	0	1
Moderate	0	0	0	0	1	0	0	0	0	1
Macrophages containing haemosiderin (Total)	0	0	0	0	0	0	1	0	0	0
Moderate	0	0	0	0	0	0	1	0	0	0
Lymph Nodes - Mediastinal										
Examined	10	9	10	7	10	8	9	10	10	10
Missing	0	1	0	3	0	1	0	0	0	0
No abnormalities detected	9	8	9	7	5	7	8	9	8	6
Sinus congestion with siderocytes (Total)	1	0	0	0	0	1	1	0	0	2
Minimal	0	0	0	0	0	1	1	0	0	2
Moderate	1	0	0	0	0	0	0	0	0	0
Sinus histiocytosis with mast cells (Total)	0	1	0	0	0	0	0	0	0	0
Moderate	0	1	0	0	0	0	0	0	0	0
Macrophages containing brown pigment (Total)	0	0	0	0	4	0	0	0	0	3
Trace	0	0	0	0	1	0	0	0	0	1
Minimal	0	0	0	0	3	0	0	0	0	2
Siderocytes (Total)	0	0	1	0	1	0	0	1	2	0
Trace	0	0	1	0	0	0	0	1	2	0
Minimal	0	0	0	0	1	0	0	0	0	0
Spleen										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	9	9	0	0	10	9	9	0	0	10

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Spleen	(Continued)									
Haemosiderosis (Total)	0	1	0	0	0	0	0	0	0	0
Minimal	0	1	0	0	0	0	0	0	0	0
Capsular inflammation (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Liver										
Examined	10	10	1	0	10	9	9	0	2	10
No abnormalities detected	7	8	0	0	8	9	8	0	1	9
Parenchymal inflammatory cells (Total)	1	2	0	0	0	0	1	0	0	1
Trace	1	0	0	0	0	0	1	0	0	1
Minimal	0	2	0	0	0	0	0	0	0	0
Portal inflammation (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Vacuolated hepatocytes at median cleft	1	0	1	0	1	0	0	0	0	0
Hepatocyte necrosis (Total)	0	0	0	0	1	0	0	0	1	0
Minimal	0	0	0	0	1	0	0	0	0	0
Marked	0	0	0	0	0	0	0	0	1	0
Pancreas										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Kidneys										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	9	3	0	0	4	8	7	0	0	7
Tubular basophilia (Total)	1	4	0	0	6	0	0	0	0	0
Trace	0	2	0	0	2	0	0	0	0	0
Minimal	1	2	0	0	4	0	0	0	0	0
Dystrophic mineralisation (Total)	0	1	0	0	0	1	1	0	0	2
Trace	0	1	0	0	0	1	0	0	0	0
Minimal	0	0	0	0	0	0	1	0	0	2

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Kidneys	(Continued)									
Dilatation of the renal pelvis (Total)	0	3	0	0	0	0	1	0	0	1
Trace	0	0	0	0	0	0	1	0	0	0
Minimal	0	3	0	0	0	0	0	0	0	1
Pelvic suburothelial inflammatory cells(Total)	0	1	0	0	0	0	0	0	0	0
Moderate	0	1	0	0	0	0	0	0	0	0
Medullary cyst	0	0	0	0	1	0	0	0	0	0
Cortical inflammatory cells (Total)	0	0	0	0	1	0	0	0	0	0
Minimal	0	0	0	0	1	0	0	0	0	0
Urinary Bladder										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	9	0	0	9	9	9	0	0	10
Serosal inflammation with siderocytes (Total)	0	1	0	0	0	0	0	0	0	0
Minimal	0	1	0	0	0	0	0	0	0	0
Urothelial hyperplasia (Total)	0	0	0	0	1	0	0	0	0	0
Minimal	0	0	0	0	1	0	0	0	0	0
Uterus										
Examined	0	0	0	0	0	9	9	1	1	10
No abnormalities detected	0	0	0	0	0	6	6	0	0	8
Luminal dilatation (Total)	0	0	0	0	0	3	3	1	1	2
Minimal	0	0	0	0	0	3	3	1	1	2
Cervix										
Examined	0	0	0	0	0	9	9	0	0	10
No abnormalities detected	0	0	0	0	0	9	9	0	0	10
Ovaries										
Examined	0	0	0	0	0	9	9	0	0	10
No abnormalities detected	0	0	0	0	0	9	9	0	0	10
Epididymides										
Examined	10	10	0	1	10	0	0	0	0	0

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Epididymides	(Continued)									
No abnormalities detected	10	10	0	1	10	0	0	0	0	0
Testes										
Examined	10	10	0	1	10	0	0	0	0	0
No abnormalities detected	8	8	0	0	9	0	0	0	0	0
Atrophy (Total)	2	2	0	1	1	0	0	0	0	0
Trace	2	0	0	0	1	0	0	0	0	0
Minimal	0	2	0	1	0	0	0	0	0	0
Thyroids										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	9	9	9	0	0	10
Ectopic thymic tissue	0	0	0	0	1	0	0	0	0	0
Parathyroids										
Examined	10	9	0	0	10	9	8	0	0	9
Missing	0	1	0	0	0	0	1	0	0	1
No abnormalities detected	10	8	0	0	10	9	8	0	0	9
Cyst	0	1	0	0	0	0	0	0	0	0
Adrenals										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Pituitary										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Salivary Glands										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Oesophagus										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Stomach										
Examined	10	10	2	0	10	9	9	2	0	10
No abnormalities detected	10	8	0	0	9	7	9	1	0	9
Focus of ectopic non-glandular epithelium within the glandular mucosa	0	2	0	0	1	1	0	0	0	1
Cysts at limiting ridge (Total)	0	0	2	0	0	1	0	1	0	0
Moderate	0	0	0	0	0	1	0	1	0	0
Duodenum										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Jejunum										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Ileum										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Caecum										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Colon										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Rectum										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Skin										
Examined	0	1	0	0	0	0	0	0	0	0

TABLE 19

(Microscopic pathology incidence summary - termination - continued)

Removal reason: Terminal	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	10	10	10	9	9	10	10	10
Skin	(Continued)									
Scab (Total)	0	1	0	0	0	0	0	0	0	0
Minimal	0	1	0	0	0	0	0	0	0	0
Eyes										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	9	10	0	0	10	9	9	0	0	10
Retinal rosettes (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Sciatic Nerve										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	9	9	9	0	0	10
Degenerate fibres (Total)	0	0	0	0	1	0	0	0	0	0
Trace	0	0	0	0	1	0	0	0	0	0
Brain										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10
Sternum										
Examined	10	10	0	0	10	9	9	0	0	10
No abnormalities detected	10	10	0	0	10	9	9	0	0	10

TABLE 20

Microscopic pathology incidence summary - withdrawal

Removal reason: Recovery	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	0	0	10	10	10	0	0	9
Pharynx										
Examined	0	0	0	0	0	1	0	0	0	0
No abnormalities detected	0	0	0	0	0	1	0	0	0	0
Lungs										
Examined	10	10	0	0	10	10	10	0	0	9
No abnormalities detected	2	0	0	0	0	6	2	0	0	0
Pneumonitis (Total)	3	9	0	0	6	2	3	0	0	3
Trace	0	1	0	0	1	2	1	0	0	2
Minimal	2	7	0	0	5	0	2	0	0	0
Moderate	1	1	0	0	0	0	0	0	0	1
Pneumonia (Total)	1	0	0	0	0	0	0	0	0	0
Minimal	1	0	0	0	0	0	0	0	0	0
Perivascular inflammatory infiltration(Total)	7	10	0	0	9	3	7	0	0	8
Trace	3	2	0	0	1	2	3	0	0	3
Minimal	2	2	0	0	3	1	1	0	0	5
Moderate	2	6	0	0	5	0	3	0	0	0
Prominent BALT (Total)	0	1	0	0	1	0	0	0	0	0
Moderate	0	1	0	0	1	0	0	0	0	0
Vascular congestion (Total)	0	1	0	0	0	0	1	0	0	0
Minimal	0	0	0	0	0	0	1	0	0	0
Aggregates of macrophages (Total)	1	1	0	0	0	0	0	0	0	0
Trace	1	1	0	0	0	0	0	0	0	0
Alveolitis with interstitial thickening of alveolar duct walls (Total)	0	0	0	0	1	0	0	0	0	1
Trace	0	0	0	0	0	0	0	0	0	1
Minimal	0	0	0	0	1	0	0	0	0	0
Brown pigment in macrophages (Total)	0	0	0	0	4	0	0	0	0	4
Trace	0	0	0	0	2	0	0	0	0	4
Minimal	0	0	0	0	2	0	0	0	0	0

TABLE 20

(Microscopic pathology incidence summary - withdrawal - continued)

Removal reason: Recovery	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	0	0	10	10	10	0	0	9
Lungs	(Continued)									
Granulomatous inflammation (Total)	1	0	0	0	1	0	0	0	0	0
Trace	0	0	0	0	1	0	0	0	0	0
Moderate	1	0	0	0	0	0	0	0	0	0
Lymph Nodes - Cervical										
Examined	1	0	0	0	0	0	0	0	0	0
Increased cellularity - generalised (Total)	1	0	0	0	0	0	0	0	0	0
Moderate	1	0	0	0	0	0	0	0	0	0
Lymph Nodes - Tracheobronchial										
Examined	10	9	0	0	10	10	9	0	0	9
Missing	0	1	0	0	0	0	1	0	0	0
No abnormalities detected	4	2	0	0	5	9	6	0	0	6
Increased lymphoid cellularity - generalised(Total)	6	7	0	0	5	1	3	0	0	3
Trace	0	0	0	0	0	1	0	0	0	0
Minimal	6	7	0	0	4	0	3	0	0	3
Moderate	0	0	0	0	1	0	0	0	0	0
Lymph Nodes - Mediastinal										
Examined	8	10	0	0	10	10	10	0	0	9
Missing	2	0	0	0	0	0	0	0	0	0
No abnormalities detected	7	10	0	0	10	9	10	0	0	7
Siderocytes (Total)	0	0	0	0	0	0	0	0	0	1
Trace	0	0	0	0	0	0	0	0	0	1
Increased lymphoid cellularity - generalised(Total)	1	0	0	0	0	0	0	0	0	1
Minimal	1	0	0	0	0	0	0	0	0	1
Sinus congestion	0	0	0	0	0	1	0	0	0	0
Liver										
Examined	1	2	0	0	0	1	0	0	0	2
No abnormalities detected	1	2	0	0	0	1	0	0	0	2

TABLE 20

(Microscopic pathology incidence summary - withdrawal - continued)

Removal reason: Recovery	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	0	0	10	10	10	0	0	9
Kidneys										
Examined	0	2	0	0	0	0	1	0	0	1
Pyelonephritis (Total)	0	0	0	0	0	0	1	0	0	0
Moderate	0	0	0	0	0	0	1	0	0	0
Dilatation of the renal pelvis (Total)	0	2	0	0	0	0	0	0	0	1
Trace	0	0	0	0	0	0	0	0	0	1
Minimal	0	2	0	0	0	0	0	0	0	0
Ureters										
Examined	0	0	0	0	0	0	1	0	0	0
No abnormalities detected	0	0	0	0	0	0	1	0	0	0
Uterus										
Examined	0	0	0	0	0	2	3	0	0	1
Luminal dilatation (Total)	0	0	0	0	0	2	3	0	0	1
Trace	0	0	0	0	0	0	1	0	0	0
Minimal	0	0	0	0	0	2	2	0	0	1
Ovaries										
Examined	0	0	0	0	0	0	0	0	0	1
Absence of corpora lutea	0	0	0	0	0	0	0	0	0	1
Epididymides										
Examined	0	0	0	0	1	0	0	0	0	0
No abnormalities detected	0	0	0	0	1	0	0	0	0	0
Pituitary										
Examined	0	0	0	0	0	1	1	0	0	0
No abnormalities detected	0	0	0	0	0	0	1	0	0	0
Hyperplasia in the pars anterior (Total)	0	0	0	0	0	1	0	0	0	0
Moderate	0	0	0	0	0	1	0	0	0	0
Stomach										
Examined	1	2	0	0	2	1	0	0	0	0

Uterus of rats 151F, Group 5 contained live foetuses

TABLE 20

(Microscopic pathology incidence summary - withdrawal - continued)

Removal reason: Recovery	Group 1	Group 2	Group 3	Group 4	Group 5	Group 1	Group 2	Group 3	Group 4	Group 5
	Males					Females				
Animals on study	20	20	10	10	20	20	20	10	10	20
Animals completed	10	10	0	0	10	10	10	0	0	9
Stomach	(Continued)									
Focus of ectopic non-glandular epithelium within the glandular mucosa	0	2	0	0	1	1	0	0	0	0
Cysts at limiting ridge (Total)	1	0	0	0	1	0	0	0	0	0
Moderate	1	0	0	0	1	0	0	0	0	0
Mammary Glands										
Examined	0	0	0	0	0	1	0	0	0	1
Mammary adenocarcinoma (Malignant)	0	0	0	0	0	1	0	0	0	1
Skin										
Examined	1	1	0	0	1	0	0	0	0	0
Scab (Total)	0	0	0	0	1	0	0	0	0	0
Minimal	0	0	0	0	1	0	0	0	0	0
Epidermal erosion (Total)	1	0	0	0	1	0	0	0	0	0
Moderate	1	0	0	0	1	0	0	0	0	0
Epidermal hyperplasia (Total)	0	1	0	0	1	0	0	0	0	0
Minimal	0	1	0	0	1	0	0	0	0	0
Tail										
Examined	0	0	0	0	1	1	0	0	0	0
Pustule	0	0	0	0	1	1	0	0	0	0
Ulceration (Total)	0	0	0	0	0	1	0	0	0	0
Minimal	0	0	0	0	0	1	0	0	0	0

APPENDIX 1

Composition and quality assurance aspects of rodent diet and water

Quality assurance aspects of rodent diet

SDS Rat and Mouse No. 1 SQC Maintenance Diet is a fixed formula diet. Each batch of diet is analysed for nutrients, possible contaminants and micro-organisms, likely to be present in the diet, and which, if in excess of specified amounts, might have an undesirable effect on the test system.

Prior to release of diet for use Huntingdon Life Sciences Quality Assurance Department checks each certificate of analysis for conformity with the specification detailed below. Occasional slight deviations to this may be permitted.

Nutrients		Calculation analysis	Acceptable range
Moisture	%	10.0	11.5% max
Crude fat	%	2.6	2.0 - 4.0
Crude protein	%	14.7	12.0 - 16.5
Crude fibre	%	5.3	2.0 - 6.0
Ash	%	5.9	3.7 - 6.2
Calcium	%	0.71	0.6 - 1.2
Phosphorus	%	0.5	0.5 - 0.75
Sodium	%	0.25	0.15 - 0.35
Chlorine	%	0.4	0.3 - 0.75
Magnesium	%	0.22	0.1 - 0.3
Potassium	%	0.66	0.5 - 0.9
Iron	mg/kg	114	100 - 300
Copper	mg/kg	11	6 - 24
Manganese	mg/kg	66	40 - 80
Zinc	mg/kg	18	30 - 50
Vitamin A	iu/kg	6303	2000 min
Vitamin E	mg/kg	75.9	35 min

Contaminants		Maximum concentration
Fluorine	mg/kg	20
Nitrates (as $NaNO_3$)	mg/kg	60
Nitrites (as $NaNO_2$)	mg/kg	5
Lead	mg/kg	2
Arsenic	mg/kg	0.5
Cadmium	mg/kg	0.5
Mercury	mg/kg	0.05
Selenium	mg/kg	0.5
Total Aflatoxins	mg/kg	5
Total PCB's	mg/kg	50
Total DDT	mg/kg	100
Dieldrin	mg/kg	20
Lindane	mg/kg	100
Heptachlor	mg/kg	10
Malathion	mg/kg	500

APPENDIX 1

(Composition and quality assurance aspects of rodent diet and water - continued)

Microbiological contents		**Maximum concentration**
Total viable organisms	per/g	5000
Mesophilic spores	per/g	5000
Salmonellae	per/g	0
Presumptive E. coli	per/g	5
E. coli type 1	per/g	0
Fungal units	per/g	200
Antibiotic activity	per/g	0

Quality assurance aspects of drinking water

The water supplied to Huntingdon Life Sciences, by Anglian Water, is potable water for human consumption. Anglian Water takes it guidelines on water quality from the EEC directive relating to water for human consumption, viz: Council Directive 80/778/EEC.

Results of routine physical and chemical examination of drinking water at source as conducted, usually weekly by the supplier, are made available to Huntingdon Life Sciences as quarterley summaries.

APPENDIX 1

Composition and Quality Assurance of SDS Rat and Mouse diet

Batch use on study

Start date	Finish date	Batch no.
24 July 1996	8 August 1996	2484
8 August 1996	3 October 1996	2660
4 October 1996	8 January 1997	2802
9 January 1997	28 January 1997	3056
29 January 1997	6 February 1997	3170

Details are stored in Huntingdon Life Sciences Archives or Quality Assurance Department

APPENDIX 2

Holding room conditions

Week beginning	Relative humidity (%)		Temperature (°C)	
	Minimum	Maximum	Minimum	Maximum
22/07/96	56	68	19.5	22.5
29/07/96	56	69	18.5	22.0
5/08/96	53	68	19.0	22.0
12/08/96	50	67	19.5	22.0
19/08/96	51	70	20.0	25.0
26/08/96	51	64	20.0	21.5
2/09/96	53	68	20.0	21.5
9/09/96	45	62	20.0	21.5
16/09/96	47	58	20.0	22.0
23/09/96	45	68	20.0	21.5
30/09/96	42	61	20.0	21.5
7/10/96	43	62	20.0	21.5
14/10/96	40	64	20.0	22.0
21/10/96	46	63	20.0	21.5
28/10/96	41	62	20.0	21.0
4/11/96	42	61	19.0	21.0
8/11/96	48	51	21.0	21.5
15/11/96	43	50	20.5	21.5
22/11/96	42	50	20.5	21.5
29/11/96	44	52	20.5	21.5
6/12/96	42	50	20.5	21.5
13/12/96	34	52	20.5	22.0
20/12/96	28	52	20.0	21.5
27/12/96	20	51	20.0	21.5
3/01/97	33	51	20.5	21.0
10/01/97	33	53	19.5	21.5
17/01/97	36	52	20.5	22.5
24/01/97	38	52	20.5	22.0
31/01/97	40	62	20.0	22.0

APPENDIX 2

(Holding room conditions - continued)

Outside limits - Temperature

Week number	Date	Duration outside limit (hours)
2	20/08/96	1

Outside limits - Humidity

Week number	Date	Duration outside limit (hours)
19	20/12/96	18
20	23/12/96	1
21	01/01/97	1
22	07/01/97	0.5
23	14/01/97	0.5
24	21/01/97	0.5
25	28/01/97	0.5

APPENDIX 3

Operating conditions of the exposure system

	1 Air Control	2 NaF	3 Low dose	4 Inter dose	5 High dose
Test substance	-	NaF	——— Cryolite ———		
Supplementary airflow (l/min)	49	49	49	49	49
Extract air flow (l/min)	60	60	60	60	60
WDF operating conditions					
Airflow to WDF (l/min)	10	10	5	10	10
Powder canister size	-	Small	Small	Small	Small
Interrupter used	No	No	Yes	Yes	No
Packing pressure (ton)	-	0.2	0.2	0.2	0.2
Packed density of powder, average (g/mm)	-	0.24	0.27	0.27	0.27
Jet used	-	Yes	Yes	Yes	Yes
Baffle size	-	Wide	Wide	Wide	Wide
Gear ratio on first exposure:[2]	-	1:0.315	1:0.07	1:0.14	0.158
Interrupter setting[1]			1s on 15s off	2s on 7s off	
Theoretical delivery (g/hour)[3]		0.151	0.0025	0.022	0.085
Theoretical aerosol concentration:[4] (mg/m^3)		41.9	0.69	6.11	23.6

WDF Wright dust feed mechanism

1 Gear ratio calculated using the interrupter setting

2 Ratio between revolutions of motor shaft and cross shaft

3 With 1:1 ratio, 30 turns of the motor shaft results in 1 mm downward movement of the container every 30 minutes, producing dispersal of xg of powder where the packed density of the powder is xg/mm

4 Theoretical concentration: $(mg/m^3) = \frac{w \times 1000000}{t \times f}$

where w = theoretical delivery of powder in 60 minutes (g)
t = duration (60 minutes)
f = chamber air flow (60 l/min)

APPENDIX 4

Chamber concentration - individual values (mg/m^3)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter dose Cryolite)	5 (High dose Cryolite)
1	1	5.2	0.22	0.92	5.5
	2	6.4	0.14	1.34	5.8
	3	6.5		1.16	5.9
	Mean	6.0	0.18	1.14	5.7
2	4	5.3	0.32	0.82	3.5
	5	5.8	0.18	0.84	3.8
	6	6.7		0.84	3.5
	Mean	5.9	0.25	0.83	3.6
3	7	5.5	0.24	1.46	4.6
	8	5.6	0.24	1.3.0	4.9
	9	5.8		1.18	4.2
	Mean	5.6	0.24	1.31	4.6
4	10	5.9	0.10	1.08	3.7
	11	5.6	0.09	1.08	4.4
	12	6.2		1.20	4.7
	Mean	5.9	0.10	1.12	4.3
5	13	5.4	0.43	1.20	4.9
	14	5.8	0.25	1.20	5.1
	15	5.7		1.10	5.4
	Mean	5.6	0.34	1.17	5.1
6	16	5.5	0.39	1.10	3.2
	17	6.3	0.34	0.84	4.2
	18	6.1		0.94	3.8
	Mean	6.0	0.37	0.96	3.7
7	19	5.2	0.34	1.22	4.5
	20	5.9	0.28	1.14	4.3
	21	5.8		1.22	4.9
	Mean	5.6	0.31	1.19	4.6
8	22	6.9	0.27	1.46	7.8
	23	7.1	0.30	1.84	7.5
	24	7.4		1.74	8.4
	Mean	7.1	0.29	1.68	7.9
9	25	6.3	0.24	0.88	2.7
	26	6.0	0.08	0.86	3.0
	27	4.8		0.80	1.9
	Mean	5.7	0.16	0.85	2.5
10	28	6.0	0.58	1.22	4.7
	29	5.7	0.37	1.34	5.0
	30	7.0		1.28	5.0
	Mean	6.2	0.48	1.28	4.9

APPENDIX 4

(Chamber concentration - individual values (mg/m^3) - continued)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter dose Cryolite)	5 (High dose Cryolite)
11	31	5.6	0.23	1.28	4.4
	32	6.9	0.16	1.24	4.5
	33	5.7		1.20	4.4
	Mean	6.1	0.20	1.24	4.4
12	34	5.7	0.23	1.34	4.8
	35	6.1	0.17	1.28	4.6
	36	6.3		1.38	4.8
	Mean	6.0	0.20	1.33	4.7
13	37	5.2	0.22	1.26	8.5
	38	5.5	0.21	1.24	4.7
	39	5.2		1.28	5.1
	Mean	5.3	0.22	1.26	6.1
14	40	5.3	0.21	1.36	5
	41	5.1	0.21	1.36	4.6
	42	5.5		1.31	4.8
	Mean	5.3	0.21	1.34	4.8
15	43	4.5	0.22	1.08	4.6
	44	5.4	0.21	1.26	5.2
	45	5.1		1.44	4.9
	Mean	5.0	0.22	1.26	4.9
16	46	5.5	0.22	1.28	4.3
	47	5.4	0.22	1.24	4.4
	48	5.7		1.28	4.7
	Mean	5.5	0.22	1.27	4.5
17	49	6.0	0.22	1.20	4.3
	50	6.4	0.23	1.36	4.6
	51	6.8		1.26	4.6
	Mean	6.4	0.23	1.27	4.5
18	52	5.0	0.24	1.24	4.3
	53	6.1	0.24	1.32	4.7
	54	6.2		1.34	4.9
	Mean	5.8	0.24	1.30	4.6
19	55	5.8	0.22	1.36	4.3
	56	5.7	0.23	1.12	4.4
	57	5.9		1.30	4.6
	Mean	5.8	0.23	1.26	4.4
20	58	5.2	0.23	1.3	4.2
	59	5.3	0.22	1.22	4.4
	60	5.5		1.24	4.5
	Mean	5.3	0.23	1.25	4.4

APPENDIX 4

(Chamber concentration - individual values (mg/m^3) - continued)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter dose Cryolite)	5 (High dose Cryolite)
21	61	5.5	0.20	1.18	4.3
	62	6.0	0.20	1.10	4.4
	63	5.4		1.18	3.5
	Mean	5.6	0.20	1.15	4.1
22	64	5.4	0.22	1.28	4.2
	65	6.0	0.22	1.28	4.7
	66	5.6		1.28	4.7
	Mean	5.7	0.22	1.28	4.5
23	67	0.2	0.30	1.22	4.7
	68	1.2	0.19	1.18	5.4
	69	5.4		1.54	4.4
	Mean	2.3	0.25	1.31	4.8
24	70	4.7	0.23	1.06	4.2
	71	5.0	0.20	1.22	4.1
	72	5.6		1.04	4.0
	Mean	5.1	0.22	1.11	4.1
25	73	3.8	0.19	1.28	3.7
	74a	0.4			
	74	3.4	0.22	1.32	4.6
	75	4.0		1.14	4.5
	75a	3.9			
	Mean	3.1	0.21	1.25	4.3
26	76	4.0	0.19	1.20	3.9
	77	4.0	0.20	1.16	4.5
	77a	5.0			
	78	4.7		1.14	4.2
	Mean	4.4	0.20	1.17	4.2
27	79	4.4	0.26	1.28	4.6
	80	6.3	0.27	1.16	5.2
	81	6.4		1.28	4.7
	Mean	5.7	0.27	1.24	4.8
28	82	0.9	0.24	1.22	3.8
	82a	4.9			
	83	6.4	0.20	1.13	3.9
	84	6.6		1.26	4.6
	Mean	4.7	0.22	1.20	4.1
29	85	5.8	0.23	1.14	4.5
	86	7.6	0.28	1.20	4.8
	87	5.4		1.36	5.2
	Mean	6.3	0.26	1.23	4.8
30	88	5.0	0.24	1.18	3.5
	89	5.0	0.22	1.00	3.9
	90	5.3		1.06	4.0
	Mean	5.1	0.23	1.08	3.8

APPENDIX 4

(Chamber concentration - individual values (mg/m^3) - continued)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter. dose Cryolite)	5 (High dose Cryolite)
31	91	5.1	0.19	0.81	4.4
	92	4.7	0.18	0.96	4.5
	93	5.5		0.88	4.3
	Mean	5.1	0.19	0.88	4.4
32	94	5.8	0.15	1.10	4.5
	95	5.8	0.15	0.96	4.8
	96	6.1		0.92	4.6
	Mean	5.9	0.15	0.99	4.6
33	97	5.1	0.13	0.78	3.9
	98	6.0	0.18	0.86	4.1
	99	6.0		0.86	4.7
	Mean	5.7	0.16	0.83	4.2
34	100	5.8	0.20	0.71	4.0
	101	6.1	0.22	0.64	4.5
	102	6.4		0.86	4.2
	Mean	6.1	0.21	0.74	4.2
35	103	6.0	0.17	1.42	4.3
	104	5.4	0.19	0.90	4.8
	105	6.1		0.98	4.4
	Mean	5.8	0.18	1.10	4.5
36	106	6.0	0.16	0.82	4.0
	107	5.8	0.22	0.98	4.5
	108	6.0		0.84	4.4
	Mean	5.9	0.19	0.88	4.3
37	109	5.6	0.18	0.88	4.6
	110	5.9	0.23	0.92	4.9
	111	6.2		1.06	5.0
	Mean	5.9	0.21	0.95	4.8
38	112	5.5	0.09	0.80	5.0
	113	5.9	0.06	1.00	4.6
	114	6.2		0.86	5.0
	Mean	5.9	0.08	0.89	4.9
39	115	5.9	0.21	1.16	4.6
	116	6.3	0.23	0.98	4.5
	117	6.2		1.02	5.2
	Mean	6.1	0.22	1.05	4.8
40	118	5.9	0.19	0.88	4.6
	119	6.1	0.20	0.74	4.2
	120	6.2		0.92	5.2
	Mean	6.1	0.20	0.85	4.7

APPENDIX 4

(Chamber concentration - individual values (mg/m^3) - continued)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter dose Cryoli te)	5 (High dose Cryolite)
41	121	6.5	0.22	0.80	4.3
	122	6.1	0.22	0.88	4.7
	123	6.5		1.04	4.8
	Mean	6.4	0.22	0.91	4.6
42	124	5.9	0.14	1.46	4.0
	125	5.5	0.14	1.06	4.6
	126	5.0		0.92	4.1
	Mean	5.5	0.14	1.15	4.2
43	127	5.4	0.18	0.56	5.9
	128	6.1	0.17	0.62	4.4
	129	6.0		0.46	1.8
	Mean	5.8	0.18	0.55	4.0
44	130	5.7	0.22	0.98	4.1
	131	5.8	0.21	0.88	4.5
	132	6.3		0.78	5.0
	Mean	5.9	0.22	0.88	4.5
45	133	6.0	0.17	0.60	4.4
	134	6.3	0.16	0.82	4.7
	135	6.7		0.80	4.3
	Mean	6.3	0.17	0.74	4.5
46	136	6.2	0.22	0.86	4.1
	137	6.4	0.23	0.88	4.3
	138	6.3		0.88	4.8
	Mean	6.3	0.23	0.87	4.4
47	139	5.8	0.18	0.78	4.4
	140	6.1	0.20	1.00	4.3
	141	7.5		0.88	4.7
	Mean	6.5	0.19	0.89	4.5
48	142	5.3	0.22	0.14	4.1
	143	6.7	0.18	0.90	4.4
	144	6.5		0.78	4.8
	Mean	6.2	0.20	0.61	4.4
49	145	5.3	0.19	0.80	4.1
	146	5.0	0.17	0.78	4.6
	147	6.4		0.98	4.9
	Mean	5.6	0.18	0.85	4.5
50	148	5.3	0.18	0.86	4.2
	149	5.4	0.22	0.86	4.8
	150	5.6		0.92	4.8
	Mean	5.4	0.20	0.88	4.6

APPENDIX 4

(Chamber concentration - individual values (mg/m^3) - continued)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter dose Cryolite)	5 (High dose Cryolite)
51	151	4.3	0.14	0.66	3.8
	152	5.7	0.14	0.86	4.8
	153	5.6		0.98	4.8
	Mean	5.2	0.14	0.83	4.5
52	154	6.1	0.03	0.86	4.1
	155	6.0	0.17	0.84	4.0
	156	5.9		0.86	4.0
	Mean	6.0	0.10	0.85	4.0
53	157	5.2	0.20	0.63	4.5
	158	4.9	0.14	0.78	5.0
	159	5.5		0.88	4.7
	Mean	5.2	0.17	0.76	4.7
54	160	6.3	0.20	0.98	4.7
	161	6.5	0.16	0.96	5.2
	162	7.6		1.06	4.8
	Mean	6.8	0.18	1.00	4.9
55	163	5.8	0.16	0.96	4.0
	164	6.1	0.20	1.10	4.2
	165	5.8		0.90	4.3
	Mean	5.9	0.18	0.99	4.2
56	166	5.8	0.18	1.06	4.8
	167	5.9	0.21	1.04	5.4
	168	6.3		1.06	5.5
	Mean	6.0	0.20	1.05	5.2
57	169	5.0	0.32	0.86	4.3
	170	5.3	0.17	0.88	4.5
	171	6.4		0.86	4.6
	Mean	5.6	0.25	0.87	4.5
58	172	6.0	0.15	0.70	4.1
	173	5.1	0.16	0.90	4.2
	174	5.5		0.88	4.0
	Mean	5.5	0.16	0.83	4.1
59	175	5.3	0.20	1.22	5.1
	176	5.3	0.25	1.12	5.5
	177	5.3		1.08	5.8
	Mean	5.3	0.23	1.14	5.5
60	178	5.4	0.15	0.82	4.2
	179	5.3	0.18	0.82	4.4
	180	5.6		0.88	4.2
	Mean	5.4	0.17	0.84	4.3

APPENDIX 4

(Chamber concentration - individual values (mg/m^3) - continued)

Exposure number	Sample	Group			
		2 (Na F)	3 (Low dose Cryolite)	4 (Inter dose Cryolite)	5 (High dose Cryolite)
61	181	5.6	0.17	1.04	4.6
	182	5.7	0.25	0.92	5.7
	183	5.7		1.02	6.0
	Mean	5.7	0.21	0.99	5.4
62	184	5.9	0.20	0.90	4.0
	185	5.4	0.21	0.82	4.5
	186	5.7		0.82	5.0
	Mean	5.7	0.21	0.85	4.5
63	187	5.3	0.20	0.86	4.1
	188	6.0	0.19	0.90	4.7
	189	5.8		0.90	4.6
	Mean	5.7	0.20	0.89	4.5
64	190	4.7	0.19	0.84	4.7
	191	5.8	0.21	0.90	4.8
	192	5.6		0.88	6.3
	Mean	5.4	0.20	0.87	5.3
65	193	6.7	0.21	0.88	4.5
	194	5.4	0.17	0.94	4.4
	195	6.2		1.30	7.1
	Mean	6.1	0.19	1.04	5.3
66	196	6.1	0.24	0.88	4.7
	197	6.4	0.21	0.90	5.6
	198	5.9		0.92	5.6
	Mean	6.1	0.23	0.90	5.3

APPENDIX 5

Individual clinical signs

Group	Animal no./sex	Observation
1 (Air control)	1M	No abnormalities detected.
	2M	No abnormalities detected.
	3M	No abnormalities detected.
	4M	No abnormalities detected.
	5M	Hair loss on head and abrasions on head Week 13.
	6M	No abnormalities detected.
	7M	No abnormalities detected.
	8M	No abnormalities detected.
	9M	No abnormalities detected.
	10M	No abnormalities detected.
	11M	Hair loss on head Week 13 to 14, 16 onwards. Abrasions on head Week 17 to 22. Hair loss on limbs Week 23 onwards.
	12M	Hair loss on head Week 13 to 14 and 17.
	13M	No abnormalities detected.
	14M	Brown staining on body Week 16.
	15M	No abnormalities detected.
	16M	Hair loss on head Week 17 and 19. Abrasions on head Week 17.
	17M	No abnormalities detected.
	18M	No abnormalities detected.
	19M	No abnormalities detected.
	20M	No abnormalities detected.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
2 (NaF)	21M	No abnormalities detected.
	22M	No abnormalities detected.
	23M	No abnormalities detected.
	24M	No abnormalities detected.
	25M	No abnormalities detected.
	26M	No abnormalities detected.
	27M	No abnormalities detected.
	28M	No abnormalities detected.
	29M	Hair loss on head Week 6 to 14. Abrasions on head Week 6 to 8 and 11 to 14.
	30M	No abnormalities detected.
	31M	Hair loss on head Week 20.
	32M	Hair loss on head Week 17, 20 to 24. Abrasions on head Week 17 and 20.
	33M	Hair loss on head Week 9, 15 to 16, 18 onwards. Abrasions on head Week 9, 15 and 22.
	34M	Hair loss on head Week 17 onwards. Abrasions on head Week 17, 20 to 22.
	35M	No abnormalities detected.
	36M	No abnormalities detected.
	37M	No abnormalities detected.
	38M	No abnormalities detected.
	39M	Red staining around left eye Week 17 to 18, 20 and 23.
	40M	Hair loss on forelimbs Week 23 onwards.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
3 (Low dose Cryolite)	41M	No abnormalities detected.
	42M	No abnormalities detected.
	43M	No abnormalities detected.
	44M	No abnormalities detected.
	45M	Hair loss on body Week 14.
	46M	No abnormalities detected.
	47M	No abnormalities detected.
	48M	No abnormalities detected.
	49M	No abnormalities detected.
	50M	No abnormalities detected.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
4 (Inter Dose Cryolite)	51M	Hair loss on head Week 14.
	52M	No abnormalities detected.
	53M	No abnormalities detected.
	54M	No abnormalities detected.
	55M	Hair loss on head Week 6 to 7 and 14. Abrasions on head Week 6 to 7.
	56M	No abnormalities detected.
	57M	No abnormalities detected.
	58M	No abnormalities detected.
	59M	No abnormalities detected.
	60M	No abnormalities detected.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
5 (High dose Cryolite)	61M	No abnormalities detected.
	62M	No abnormalities detected.
	63M	No abnormalities detected.
	64M	No abnormalities detected.
	65M	No abnormalities detected.
	66M	No abnormalities detected.
	67M	No abnormalities detected.
	68M	No abnormalities detected.
	69M	No abnormalities detected.
	70M	No abnormalities detected.
	71M	No abnormalities detected.
	72M	No abnormalities detected.
	73M	No abnormalities detected.
	74M	No abnormalities detected.
	75M	No abnormalities detected.
	76M	Hair loss on head Week 18, 20, 23 onwards. Abrasions on head Week 23 to 26.
	77M	Hair loss on body Week 6.
	78M	Hair loss and abrasions on head Week 23 onwards.
	79M	Hair loss on head Week 15. Abrasions on body Week 17 onwards. Weeping area, lumber region Week 20 to 22. Isolated Week 17 onwards.
	80M	No abnormalities detected.

APPENDIX 5

Individual clinical signs

Group	Animal no./sex	Observation
1 (Air Control)	81F	No abnormalities detected.
	82F	No abnormalities detected.
	83F	No abnormalities detected.
	84F	Died Week 14.
	85F	No abnormalities detected.
	86F	No abnormalities detected.
	87F	No abnormalities detected.
	88F	Hair loss on body Week 14.
	89F	Hair loss on head Week 10 to 14. Hair loss on body Week 11 to 14.
	90F	No abnormalities detected.
	91F	No abnormalities detected.
	92F	No abnormalities detected.
	93F	No abnormalities detected.
	94F	Brown staining on head Week 20 onwards.
	95F	No abnormalities detected.
	96F	No abnormalities detected.
	97F	Brown staining on body Week 15 and 17 to 22.
	98F	Hair loss on body Week 14. Hair loss on head Week 14 to 15 and 17 onwards. Mass right abdominal region Week 20 onwards. Mass (10 x 12 mm) Week 20; (24 x 20 mm) Week 24; (20 x 10 mm) Week 27.
	99F	Hair loss limbs Week 14 to 15 and 17 to 22. Hair loss on head Week 17 to 18. Hair loss on forelimbs and brown staining on head Week 23 onwards.
	100F	Hair loss on head Week 18 to 19.

APPENDIX 5

Individual clinical signs

Group	Animal no./sex	Observation
2 (NaF)	101F	No abnormalities detected.
	102F	Hair loss on body Week 8 to 14.
	103F	Hair loss on body Week 14.
	104F	No abnormalities detected.
	105F	Hair loss on body Week 8 to 14.
	106F	Died Week 6.
	107F	No abnormalities detected.
	108F	No abnormalities detected.
	109F	No abnormalities detected.
	110F	No abnormalities detected.
	111F	Brown staining on body Week 9 to 13.
	112F	Hair loss on body Week 20 onwards.
	113F	Brown staining on head Week 27.
	114F	Brown staining on body Week 9 to 13 and 15. Hair loss on body Week 23 onwards.
	115F	Brown staining on body Week 3 to 13 and 15. Brown staining on head Week 16 to 27. Hair loss on body Week 20 onwards.
	116F	Brown staining on body Week 11 to 13.
	117F	Brown staining on body Week 9 to 13, 15, 17 to 19.
	118F	No abnormalities detected.
	119F	Brown staining on body Week 9 to 13, 15, 17 to 19.
	120F	No abnormalities detected.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
3 (Low dose Cryolite)	121F	No abnormalities detected.
	122F	No abnormalities detected.
	123F	No abnormalities detected.
	124F	No abnormalities detected.
	125F	No abnormalities detected.
	126F	Hair loss on limbs Week 9 to 14. Hair loss on body Week 10 to 14.
	127F	Hair loss on body Week 7 and 11 to 14.
	128F	No abnormalities detected.
	129F	Hair loss on body Week 11 to 14.
	130F	Hair loss on body Week 11 to 14.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
4 (Inter dose Cryolite)	131F	No abnormalities detected.
	132F	No abnormalities detected.
	133F	No abnormalities detected.
	134F	No abnormalities detected.
	135F	No abnormalities detected.
	136F	No abnormalities detected.
	137F	Hair loss on body Week 7.
	138F	No abnormalities detected.
	139F	Hair loss on body Week 10 to 14.
	140F	No abnormalities detected.

APPENDIX 5

(Individual clinical signs - continued)

Group	Animal no./sex	Observation
5 (High dose Cryolite)	141F	No abnormalities detected.
	142F	No abnormalities detected.
	143F	No abnormalities detected.
	144F	Brown staining on head Week 7.
	145F	No abnormalities detected.
	146F	No abnormalities detected.
	147F	No abnormalities detected.
	148F	No abnormalities detected.
	149F	No abnormalities detected.
	150F	No abnormalities detected.
	151F	No abnormalities detected.
	152F	No abnormalities detected.
	153F	Masses upper right thoracic region Week 12 onwards. Mass A (10 x 10 mm) Week 12; (18 x 14 mm) Week 20; (13 x 19 mm) Week 27. Mass B (5 x 5 mm) Week 12; (24 x 15 mm) Week 22; (22 x 14 mm) Week 27. Hair loss on body Week 14 and 17.
	154F	No abnormalities detected.
	155F	Hair loss on body Week 4 to 19, 21 to 22, 24 onwards.
	156F	Hair loss on body Week 11 to 14. Hair loss on head Week 25 onwards.
	157F	Hair loss on body Week 11 onwards. Brown staining on body Week 15. Hair loss on head Week 25 onwards.
	158F	Brown staining on body Week 11 to 19. Hair loss on body Week 11 to 14. Hair loss on head Week 23 onwards.
	159F	Hair loss on body Week 10 onwards. Hair loss on head Week 23 onwards.
	160F	No abnormalities detected.

APPENDIX 6

Bodyweights - individual values (g)

Group 1M: Air Control

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
1	1	195	257	302	327	356	378	403	416	438	444	457	464	474	476
	2	193	236	242	287	274	289	305	323	338	350	358	360	365	379
	3	208	261	272	297	315	334	353	386	403	425	432	447	460	457
	4	212	266	276	296	306	329	352	375	394	416	435	452	448	453
	5	205	265	287	302	330	343	362	372	382	396	405	412	427	429
2	6	209	252	281	306	355	354	366	383	397	398	409	417	421	423
	7	193	249	260	279	288	307	320	332	343	360	370	385	398	397
	8	205	267	302	333	363	385	401	414	431	438	461	470	478	482
	9	226	287	310	340	367	398	416	426	440	460	476	484	496	495
	10	204	257	278	297	309	328	350	358	370	383	389	391	394	403
3	11	216	267	283	309	340	363	381	397	407	432	458	470	486	486
	12	203	256	281	305	323	336	339	353	369	383	403	417	425	433
	13	192	247	282	310	325	350	362	369	393	409	422	433	442	447
	14	205	250	267	288	307	329	350	367	389	408	424	438	450	451
	15	204	252	279	308	327	345	371	386	403	416	430	445	450	456
4	16	215	273	297	324	345	355	373	382	392	403	421	435	448	446
	17	221	284	323	354	380	408	427	440	459	471	496	509	518	523
	18	203	260	290	318	340	357	374	384	398	418	422	422	433	445
	19	190	247	271	303	328	337	354	369	385	400	408	411	427	434
	20	193	257	268	280	298	316	342	366	386	407	428	443	461	469

APPENDIX 6

(Bodyweights - continued)

Group 1M: Air Control

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
1	1	475													
	2	380													
	3	448													
	4	442													
	5	419													
2	6	421													
	7	385													
	8	475													
	9	494													
	10	402													
3	11	471	482	501	520	532	543	552	556	573	581	594	605	619	614
	12	426	438	452	464	499	490	497	507	510	527	529	531	540	533
	13	439	454	470	481	484	516	526	532	545	556	574	579	586	594
	14	445	454	467	484	483	494	497	502	507	512	528	530	537	533
	15	459	486	503	517	529	543	540	551	560	571	578	581	588	587
4	16	442	466	499	513	524	537	545	563	566	582	598	601	605	608
	17	530	556	575	598	609	618	629	639	646	660	680	684	697	690
	18	447	472	493	504	517	522	534	552	565	573	584	591	597	598
	19	440	463	479	492	504	516	527	535	548	559	568	571	584	580
	20	457	481	517	532	548	561	573	585	596	609	621	625	636	644

APPENDIX 6

(Bodyweights - continued)

Group 2M: NaF

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
5	21	202	262	288	306	334	357	376	376	400	411	430	444	457	466
	22	186	228	239	259	279	292	301	318	325	336	340	350	351	357
	23	186	236	261	285	310	333	351	355	371	385	400	406	416	410
	24	213	274	303	328	353	373	387	403	422	443	455	458	471	471
	25	216	278	308	332	360	383	413	446	472	494	502	525	539	553
6	26	195	253	284	317	350	367	379	402	418	430	441	454	462	464
	27	205	254	268	288	308	326	336	353	362	379	385	407	411	420
	28	195	254	284	319	339	356	369	387	399	412	424	438	444	446
	29	216	274	295	328	356	383	394	414	425	446	456	462	481	488
	30	196	245	256	289	307	329	356	381	397	402	420	430	435	447
7	31	207	271	298	325	349	366	385	403	418	435	447	457	469	471
	32	201	250	261	278	322	304	307	317	331	340	352	367	368	373
	33	218	273	284	300	300	339	355	373	388	403	418	436	441	445
	34	221	283	305	334	356	367	382	407	416	426	444	458	472	477
	35	195	254	289	320	344	369	385	400	409	418	418	435	444	445
8	36	197	257	286	322	354	382	397	405	413	423	439	455	458	474
	37	205	259	286	318	341	368	385	399	415	430	441	454	464	470
	38	222	284	326	369	394	420	439	468	477	508	520	537	551	557
	39	192	246	274	305	332	354	365	381	392	406	419	433	434	443
	40	217	274	288	306	331	359	387	407	421	449	463	470	478	493

APPENDIX 6

(Bodyweights - continued)

Group 2M: NaF

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
5	21	458													
	22	362													
	23	397													
	24	462													
	25	553													
6	26	456													
	27	413													
	28	441													
	29	491													
	30	438													
7	31	467	501	526	534	553	578	590	593	605	616	626	633	643	643
	32	379	390	396	405	413	425	424	427	436	442	443	444	455	455
	33	440	460	484	498	518	525	543	548	559	568	590	594	609	607
	34	475	493	516	537	549	564	576	581	592	611	616	630	641	640
	35	441	468	483	500	513	522	544	549	564	569	575	577	590	598
8	36	478	499	517	529	545	550	557	573	579	582	595	597	600	601
	37	467	490	516	531	542	560	571	576	589	602	613	617	633	620
	38	567	584	603	625	634	640	660	686	675	697	710	720	737	746
	39	438	449	469	480	487	496	509	512	519	528	535	545	555	554
	40	499	518	545	562	563	582	589	606	614	627	644	650	657	666

APPENDIX 6

(Bodyweights - continued)

Group 3M: Low dose

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
9	41	199	254	280	311	339	362	381	397	404	411	421	423	421	417
	42	188	245	271	291	313	328	351	367	381	398	408	415	428	423
	43	213	281	304	333	358	387	398	417	431	442	461	470	475	448
	44	212	266	296	310	334	353	373	388	413	425	436	449	463	464
	45	218	272	283	305	332	363	384	407	420	433	455	466	477	483
10	46	198	247	271	277	302	317	330	352	364	378	379	386	392	406
	47	202	244	256	273	282	297	313	327	333	344	351	362	361	367
	48	199	245	258	284	312	318	328	377	376	392	409	409	425	428
	49	224	294	338	380	424	458	489	520	546	555	567	570	583	585
	50	206	253	281	296	310	332	350	367	377	395	408	400	420	424

APPENDIX 6

(Bodyweights - continued)

Group 3M: Low dose

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
9	41	414													
	42	420													
	43	464													
	44	456													
	45	475													
10	46	409													
	47	370													
	48	427													
	49	580													
	50	418													

APPENDIX 6

(Bodyweights - continued)

Group 4M: Inter dose

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
11	51	212	275	301	329	356	383	406	428	445	459	476	491	497	500
	52	202	252	277	308	332	351	372	394	410	423	435	444	449	456
	53	220	287	307	336	361	384	403	427	436	438	463	474	466	471
	54	205	265	298	329	353	375	397	412	425	426	436	447	453	452
	55	219	274	292	311	325	343	361	382	401	418	423	436	443	446
12	56	194	251	297	303	332	357	377	399	412	408	425	429	434	446
	57	207	261	266	281	296	317	338	356	364	375	391	401	408	416
	58	214	266	288	307	328	349	361	382	387	409	426	433	451	452
	59	196	253	270	292	307	326	349	366	395	399	414	421	429	434
	60	215	268	282	292	315	332	345	368	396	412	431	436	445	462

APPENDIX 6

(Bodyweights - continued)

Group 4M: Inter dose

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
11	51	491													
	52	443													
	53	470													
	54	456													
	55	438													
12	56	443													
	57	413													
	58	439													
	59	424													
	60	465													

APPENDIX 6

(Bodyweights - continued)

Group 5M: High dose

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
13	61	196	247	269	298	316	326	353	377	392	405	419	437	447	461
	62	204	258	275	297	313	324	331	337	353	359	365	364	370	363
	63	215	271	282	289	300	311	331	350	362	376	365	373	388	396
	64	197	241	268	298	318	342	364	373	384	395	404	407	415	425
	65	209	279	318	354	377	405	426	453	481	499	512	524	535	535
14	66	210	260	286	307	334	347	365	383	395	413	393	437	442	460
	67	212	266	287	308	320	338	364	382	391	401	404	416	425	432
	68	200	251	268	290	305	326	340	351	356	377	385	392	399	399
	69	194	245	270	289	312	334	351	378	391	413	428	442	457	462
	70	194	249	283	314	336	349	356	375	383	398	402	414	419	423
15	71	194	239	254	267	281	298	315	331	337	352	362	365	371	383
	72	205	260	280	303	316	326	334	351	366	377	383	395	407	410
	73	199	256	278	306	331	358	379	396	411	417	423	432	442	452
	74	220	276	300	315	324	344	367	384	411	419	432	444	451	451
	75	199	256	296	333	364	388	413	445	457	470	491	499	508	499
16	76	191	244	256	271	288	306	319	320	330	337	349	355	365	365
	77	201	252	260	285	303	321	341	363	382	396	400	410	427	415
	78	214	261	274	299	323	342	359	383	395	405	419	439	445	442
	79	210	274	293	319	353	372	389	403	413	423	430	435	438	421
	80	207	255	263	268	282	297	316	326	336	352	364	381	387	381

APPENDIX 6

(Bodyweights - continued)

Group 5M: High dose

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
13	61	454													
	62	360													
	63	384													
	64	419													
	65	523													
14	66	453													
	67	424													
	68	397													
	69	458													
	70	419													
15	71	382	405	421	437	447	455	467	474	480	483	497	504	512	505
	72	402	439	449	466	473	483	499	509	513	519	527	534	544	534
	73	444	478	494	509	526	538	541	559	571	583	591	595	603	598
	74	449	474	488	511	520	526	541	552	562	558	570	567	574	557
	75	503	528	551	563	573	595	603	606	619	626	639	642	652	647
16	76	375	397	418	422	437	447	458	459	471	483	498	504	522	531
	77	425	436	459	479	492	496	501	510	516	519	532	541	555	555
	78	459	490	518	532	543	549	561	571	572	576	584	591	598	599
	79	425	450	470	484	497	500	509	506	522	530	542	548	560	559
	80	384	409	422	445	449	457	470	475	491	500	515	524	530	532

APPENDIX 6

(Bodyweights - continued)

Group 1F: Air Control

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
17	81	162	173	180	188	195	194	207	218	220	224	228	235	237	236
	82	173	194	219	224	227	227	241	244	245	239	250	248	253	249
	83	172	194	213	220	226	231	240	250	254	253	257	257	256	257
	84	173	198	206	217	231	239	247	251	253	253	258	260	264	273
	85	190	213	232	242	251	246	265	268	271	267	279	283	284	283
18	86	175	199	203	202	226	231	233	236	245	244	246	251	258	258
	87	173	191	205	215	219	227	237	248	252	251	261	271	265	261
	88	178	207	216	222	229	245	252	260	261	262	266	272	272	279
	89	184	198	202	217	229	236	240	253	262	266	268	277	284	284
	90	184	215	224	233	244	263	268	278	276	285	293	291	289	298
19	91	178	201	213	226	239	243	258	265	272	274	279	288	292	298
	92	175	203	217	227	237	245	257	257	261	260	273	273	281	277
	93	171	195	207	211	238	229	235	242	252	252	252	254	262	258
	94	185	214	224	235	246	256	259	263	273	279	283	288	307	297
	95	188	212	219	227	232	244	247	258	259	267	270	272	273	271
20	96	188	209	213	220	240	247	259	264	279	277	281	277	291	290
	97	176	195	204	215	228	229	237	244	257	252	254	260	264	259
	98	197	228	229	244	248	246	261	254	271	283	277	273	288	278
	99	167	187	204	210	215	228	235	238	236	251	253	253	243	249
	100	164	188	194	200	203	209	213	220	222	226	228	237	238	242

APPENDIX 6

(Bodyweights - continued)

Group 1F: Air Control

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
17	81	234													
	82	248													
	83	259													
	84														
	85	284													
18	86	256													
	87	261													
	88	276													
	89	271													
	90	293													
19	91	294	309	321	325	326	336	339	334	341	350	372	391	384	395
	92	277	291	295	306	308	309	310	315	317	312	317	319	318	321
	93	257	264	276	285	291	292	299	303	307	308	313	321	325	320
	94	300	288	310	310	314	321	325	324	326	330	326	334	339	340
	95	271	286	289	291	309	310	316	327	339	337	340	350	342	343
20	96	291	298	315	323	326	329	338	342	344	343	346	352	354	346
	97	265	277	280	275	277	283	282	276	288	290	293	298	308	302
	98	276	291	298	299	304	310	313	318	319	323	326	333	334	338
	99	250	256	256	268	274	275	277	285	283	292	281	294	299	291
	100	231	245	245	248	255	263	262	257	263	267	267	267	270	266

APPENDIX 6

(Bodyweights - continued)

Group 2F: NaF

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
21	101	174	202	206	217	239	241	242	258	265	262	258	266	272	267
	102	181	211	219	223	241	255	263	273	271	277	285	286	284	293
	103	161	186	190	193	203	209	213	210	228	228	226	223	235	232
	104	174	191	206	223	229	225	242	248	249	241	249	252	251	251
	105	184	208	207	227	242	253	250	255	256	262	261	264	268	266
22	106	170	186	186	196	201	202	210							
	107	174	192	202	199	218	226	231	232	244	241	239	245	248	245
	108	177	203	217	231	233	254	257	272	270	285	289	293	291	292
	109	181	216	223	233	244	254	258	265	266	269	265	266	273	273
	110	174	204	216	225	224	240	248	254	255	264	270	266	267	277
23	111	175	207	222	230	235	246	251	251	254	265	272	273	266	273
	112	178	198	207	216	211	226	227	237	239	241	243	249	242	248
	113	186	211	212	233	241	247	241	263	266	272	273	288	287	284
	114	171	191	194	208	219	226	231	242	251	258	254	263	266	269
	115	165	182	191	199	209	218	226	233	235	245	253	273	264	262
24	116	169	198	209	212	222	231	240	246	255	258	260	261	270	269
	117	165	195	199	211	211	225	227	228	226	237	238	235	235	239
	118	171	199	205	219	225	238	249	254	257	269	275	270	274	281
	119	171	193	199	211	217	226	233	234	240	245	247	248	254	254
	120	176	211	219	229	237	245	249	247	256	260	260	257	270	266

APPENDIX 6

(Bodyweights - continued)

Group 2F: NaF

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
21	101	256													
	102	288													
	103	229													
	104	251													
	105	258													
22	106														
	107	253													
	108	288													
	109	271													
	110	267													
23	111	271	294	292	308	308	314	304	303	312	315	315	323	324	316
	112	245	248	253	269	274	276	272	278	275	277	281	286	290	289
	113	275	299	314	318	321	332	339	350	351	359	362	370	373	374
	114	262	275	288	289	296	303	303	310	314	316	316	316	310	315
	115	261	275	276	276	278	280	284	288	289	289	290	292	293	295
24	116	270	280	294	296	298	306	308	305	312	315	321	315	323	331
	117	240	252	251	260	270	264	259	269	272	277	276	281	277	279
	118	280	300	300	308	315	318	315	326	327	331	333	334	344	344
	119	255	271	270	279	280	285	289	296	297	305	312	313	319	318
	120	264	270	286	293	292	294	297	305	304	304	310	316	315	312

APPENDIX 6

(Bodyweights - continued)

Group 3F: Low dose

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
25	121	176	186	186	196	206	212	217	230	237	241	237	248	253	251
	122	168	192	195	206	215	221	230	242	246	255	259	261	262	267
	123	187	208	213	228	239	240	249	264	269	263	269	278	280	278
	124	185	215	228	234	256	263	262	268	283	287	294	283	293	297
	125	185	200	215	223	227	251	267	285	287	292	301	302	305	313
26	126	179	203	208	217	231	230	242	259	261	255	272	265	275	278
	127	183	192	194	207	206	210	221	230	233	234	241	241	240	240
	128	172	191	195	199	208	218	227	226	232	236	241	236	244	250
	129	169	180	192	204	209	206	221	229	231	223	234	236	242	232
	130	173	199	207	204	220	229	232	231	242	243	241	238	250	251

APPENDIX 6

(Bodyweights - continued)

Group 3F: Low dose

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
25	121	238													
	122	265													
	123	273													
	124	283													
	125	304													
26	126	269													
	127	236													
	128	246													
	129	230													
	130	248													

APPENDIX 6

(Bodyweights - continued)

Group 4F: Inter dose

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
27	131	184	205	209	221	230	231	232	244	251	255	248	256	261	259
	132	166	183	187	187	201	210	212	213	219	225	222	225	231	230
	133	165	184	188	192	194	201	210	217	221	228	229	230	227	230
	134	165	176	181	180	188	197	204	207	206	211	213	211	216	218
	135	203	235	242	240	262	276	291	297	308	323	325	328	340	329
28	136	162	170	187	194	202	206	215	220	225	226	232	236	241	239
	137	180	198	208	217	226	233	231	241	249	253	245	257	258	260
	138	179	205	219	232	234	250	262	269	263	275	270	278	280	282
	139	181	204	207	208	222	231	231	234	245	249	245	250	258	256
	140	180	209	221	228	245	255	250	255	261	273	271	277	286	290

APPENDIX 6

(Bodyweights - continued)

Group 4F: Inter dose

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
27	131	260													
	132	236													
	133	225													
	134	218													
	135	324													
28	136	234													
	137	248													
	138	283													
	139	248													
	140	282													

APPENDIX 6

(Bodyweights - continued)

Group 5F: High dose

Cage number	Animal number	Week													
		-1	0	1	2	3	4	5	6	7	8	9	10	11	12
29	141	161	179	184	185	190	196	203	206	211	217	218	223	230	232
	142	181	206	207	220	222	227	228	235	244	250	247	255	262	262
	143	191	218	236	249	257	263	272	283	283	279	288	295	301	298
	144	168	192	194	200	207	217	223	240	239	245	241	249	249	254
	145	170	189	201	204	205	207	215	221	229	222	230	231	231	229
30	146	183	207	215	223	236	241	243	242	245	252	258	247	260	262
	147	183	214	230	241	257	270	269	262	274	273	275	268	281	291
	148	179	205	214	224	234	244	251	259	263	271	278	277	283	284
	149	164	186	195	202	213	220	229	226	238	239	244	238	238	245
	150	161	177	178	192	205	208	211	216	221	224	225	231	232	235
31	151	173	192	204	219	231	236	236	252	257	256	258	(285)	(297)	(309)
	152	183	195	202	210	222	230	240	243	253	257	260	(263)	(269)	(270)
	153	176	194	214	229	239	234	244	250	258	256	258	(269)	(266)	(256)
	154	177	192	202	219	230	231	242	247	258	262	268	(273)	(271)	(273)
	155	178	200	209	203	226	231	238	234	244	246	247	(276)	(268)	(261)
32	156	170	196	208	220	229	237	235	242	251	244	247	256	256	252
	157	180	205	203	212	228	236	241	245	254	261	263	262	267	269
	158	184	209	207	223	234	238	232	239	248	253	249	256	264	260
	159	177	206	214	227	227	252	254	253	248	263	267	266	251	268
	160	159	177	192	205	213	218	232	238	249	245	254	248	258	267

( ) Value excluded from means and statistical analysis. Accidental inclusion in Cage 31 of a male rat overnight 10/11 October (week 10)

APPENDIX 6

(Bodyweights - continued)

Group 5F: High dose

Cage number	Animal number	Week													
		13	14	15	16	17	18	19	20	21	22	23	24	25	26
29	141	229													
	142	257													
	143	297													
	144	249													
	145	229													
30	146	258													
	147	285													
	148	278													
	149	241													
	150	225													
31	151														
	152	(266)	276	291	294	296	297	302	308	308	309	312	321	313	314
	153	(265)	284	282	300	308	310	302	315	320	318	321	326	330	332
	154	(272)	296	305	313	326	335	325	343	351	343	344	351	351	342
	155	(254)	282	300	309	308	319	330	324	321	332	344	341	337	337
32	156	250	270	273	287	289	294	288	298	303	305	304	308	311	310
	157	262	272	287	295	302	301	313	316	312	315	324	326	326	318
	158	257	278	288	293	290	307	313	313	307	310	316	316	313	316
	159	267	279	296	298	293	303	311	312	304	320	320	321	321	316
	160	261	277	290	305	307	320	319	327	339	333	344	344	342	334

APPENDIX 7

Food consumption - cage mean values (g/rat/week)

Week	Group 1M: Air Control				Group 2M: NaF				Group 3M: Low dose	
	Cage									
	1	2	3	4	5	6	7	8	9	10
-1	207	207	215	218	209	207	212	208	214	205
1	189	189	204	207	200	192	202	199	195	193
2	177	182	189	198	187	194	193	208	198	193
3	187	188	201	203	202	197	201	207	205	199
4	187	192	201	199	212	209	197	221	207	205
5	192	192	202	207	209	200	199	220	210	207
6	201	190	208	216	208	210	202	224	219	217
7	201	187	209	214	216	210	207	217	218	209
8	199	188	212	214	205	205	199	217	215	211
9	197	190	216	217	212	208	206	218	214	205
10	197	191	210	212	210	207	197	223	218	199
11	192	186	209	211	214	201	199	218	203	206
12	191	188	207	215	209	211	198	226	203	205
13	164	161	179	193	183	177	185	202	179	179
14			211	222			213	228		
15			215	233			220	232		
16			212	225			218	218		
17			213	223			219	223		
18			217	222			218	224		
19			212	219			220	228		
20			212	224			216	224		
21			210	221			214	221		
22			214	222			211	222		
23			220	231			218	236		
24			214	221			215	232		
25			213	219			214	229		
26			192	199			195	202		

APPENDIX 7

(Food consumption - continued)

Week	Group 4M: Inter dose				Group 5M: High dose	
	Cage					
	11	12	13	14	15	16
-1	219	221	208	203	204	213
1	204	198	194	185	187	180
2	202	181	195	188	183	192
3	201	188	193	190	188	188
4	211	198	194	199	189	201
5	210	197	199	200	196	200
6	219	204	204	206	197	203
7	221	210	212	208	198	200
8	209	198	205	203	194	199
9	214	208	201	194	198	198
10	211	200	201	201	197	199
11	201	202	201	195	195	197
12	206	204	196	195	192	197
13	176	184	173	171	178	183
14					201	207
15					211	214
16					211	211
17					210	211
18					206	208
19					212	206
20					210	209
21					206	210
22					202	211
23					211	222
24					205	217
25					204	219
26					180	197

APPENDIX 7

(Food consumption - continued)

Week	Group 1F: Air control				Group 2F: NaF				Group 3F: Low dose	
	Cage									
	17	18	19	20	21	22	23	24	25	26
-1	151	160	156	164	166	149	159	150	154	147
1	150	154	148	149	139	143	147	141	148	138
2	150	153	150	150	145	145	146	147	149	138
3	149	159	152	153	146	146	148	148	152	140
4	154	169	154	156	141	156	155	149	155	142
5	154	163	156	158	150	151	152	151	160	150
6	154	167	156	161	150	156	158	152	168	149
7	149	165	154	163	146	154	155	151	167	152
8	146	158	151	159	138	154	155	146	156	156
9	153	159	152	160	146	150	152	147	156	148
10	146	164	149	158	141	152	159	142	154	141
11	144	158	151	156	140	149	148	146	152	143
12	145	161	145	149	143	156	149	147	157	143
13	130	140	134	140	125	131	139	137	130	126
14			156	162			162	157		
15			159	163			162	158		
16			159	161			161	152		
17			158	164			160	146		
18			154	168			159	148		
19			155	164			154	146		
20			153	167			158	149		
21			155	163			157	146		
22			152	161			154	146		
23			159	171			163	155		
24			167	173			159	148		
25			157	168			158	152		
26			150	152			144	144		

APPENDIX 7

(Food consumption - continued)

Week	Group 4F: Inter dose			Group 5F: Low dose		
	Cage					
	27	28	29	30	31	32
-1	154	157	152	157	146	155
1	145	150	141	147	142	148
2	146	152	142	152	146	154
3	151	157	143	155	149	155
4	155	163	146	152	151	161
5	157	159	148	152	154	160
6	162	165	152	157	156	163
7	157	159	151	154	156	165
8	154	160	150	149	152	159
9	149	160	151	149	152	160
10	148	158	148	147	(160)	158
11	149	159	149	146	(152)	157
12	147	156	146	147	(129)	157
13	127	136	130	128	(128)	142
14					170	169
15					175	175
16					170	173
17					169	169
18					164	173
19					158	172
20					162	170
21					158	167
22					150	161
23					160	173
24					153	164
25					151	165
26					145	148

( ) Value excluded from means and statistical analysis

APPENDIX 8

Water consumption - cage mean values (g/rat/week)

Week	Group 1M: Air control				Group 2M: NaF				Group 3M: Low dose	
	Cage									
	1	2	3	4	5	6	7	8	9	10
-1	211.4	214.8	227.4	213.0	204.0	229.6	221.0	216.6	212.4	211.4
1	214.4	210.0	213.4	219.0	205.8	224.2	220.8	215.2	223.2	220.2
2	208.0	205.0	202.6	199.4	211.4	220.8	211.0	217.8	218.0	206.8
3	210.8	208.8	205.8	202.0	210.6	223.0	215.6	215.2	224.0	215.0
4	212.6	206.0	217.6	202.8	220.6	231.2	213.2	233.2	237.4	216.2
5	216.4	206.6	225.8	207.6	229.8	241.2	216.0	236.2	228.2	223.0
6	233.8	208.2	227.4	216.8	225.4	246.6	213.4	233.2	235.8	231.8
7	243.4	214.2	234.8	226.2	235.6	259.4	227.8	249.4	250.2	228.2
8	237.0	202.2	228.6	217.4	224.6	236.0	217.2	260.4	239.6	226.0
9	242.4	218.4	237.4	218.2	235.4	242.4	225.6	266.0	250.2	230.4
10	248.2	223.4	244.0	223.4	233.4	252.2	220.2	281.2	249.8	236.0
11	251.4	225.8	253.0	226.8	239.8	256.8	234.0	272.6	255.4	234.8
12	246.8	226.6	254.0	226.2	237.8	253.8	221.6	274.8	272.6	241.4
13	230.6	202.6	247.2	241.0	225.8	240.2	245.2	285.6	234.4	219.6
14			258.4	220.6			238.0	264.6		
15			250.4	232.2			241.0	257.8		
16			247.4	226.8			239.6	249.0		
17			229.4	219.0			236.2	251.4		
18			244.4	218.4			225.8	264.8		
19			249.8	219.6			221.6	266.6		
20			256.6	231.4			219.0	252.2		
21			255.8	226.4			219.6	262.8		
22			242.2	213.0			206.0	251.6		
23			233.4	217.2			208.4	238.6		
24			234.8	211.2			219.6	229.8		
25			242.0	215.2			210.4	248.8		
26			205.0	190.2			187.0	233.6		

APPENDIX 8

(Water consumption - continued)

Week	Group 4M: Inter dose			Group 5M: High dose		
	Cage					
	11	12	13	14	15	16
-1	236.2	220.6	218.4	216.6	210.8	225.6
1	236.2	214.4	213.4	222.6	197.6	203.8
2	211.2	190.0	210.8	211.8	183.2	204.0
3	211.0	189.2	206.0	209.8	182.6	214.4
4	217.2	190.6	212.4	216.6	188.4	226.6
5	218.4	196.2	229.6	218.0	191.4	230.6
6	220.4	203.6	231.2	229.6	206.0	239.2
7	221.0	221.6	240.2	233.4	210.0	245.0
8	221.8	208.6	244.6	233.8	205.6	240.8
9	232.8	219.2	244.0	228.6	205.8	247.4
10	228.4	224.0	246.4	240.4	215.0	245.6
11	232.4	223.8	268.8	212.8	215.8	261.8
12	228.0	235.6	262.8	214.2	219.0	(334.0)
13	211.4	212.0	228.0	201.4	230.8	275.8
14					225.6	254.2
15					230.6	251.2
16					238.2	252.6
17					227.6	245.0
18					217.4	245.8
19					221.6	246.2
20					225.2	252.4
21					217.8	251.2
22					211.8	242.2
23					207.6	228.0
24					196.0	235.6
25					197.8	230.0
26					184.8	221.0

( ) Value excluded from means and statistical analysis

APPENDIX 8

(Water consumption - continued)

Week	Group 1F: Air control				Group 2F: NaF				Group 3F: Low dose	
	Cage									
	17	18	19	20	21	22	23	24	25	26
-1	149.6	177.0	168.8	173.8	156.0	155.4	174.6	164.6	162.2	167.2
1	167.2	178.4	174.2	173.2	155.4	160.6	178.6	168.4	166.6	169.4
2	162.0	173.8	168.4	172.4	162.2	163.8	184.0	173.0	166.6	173.0
3	163.8	178.6	164.6	174.6	166.2	165.8	182.2	175.0	164.8	214.2
4	156.6	183.0	172.4	175.6	167.2	167.0	188.4	176.8	167.0	230.2
5	169.2	184.2	173.0	183.6	174.6	169.4	190.8	176.6	176.0	219.4
6	169.4	192.2	169.0	183.8	173.2	175.0	191.0	179.8	175.2	252.8
7	166.6	191.6	178.6	189.8	175.4	180.5	203.8	178.6	183.8	232.4
8	157.0	186.0	170.4	179.8	181.1	171.3	192.6	174.6	184.0	185.0
9	180.2	193.2	173.8	182.4	185.0	182.8	202.4	180.0	197.0	207.4
10	176.2	205.6	177.6	188.0	179.8	173.8	216.8	189.2	193.8	193.2
11	194.4	204.4	195.6	194.0	181.2	175.8	210.2	202.8	198.0	219.2
12	184.4	213.6	185.6	184.0	210.8	180.0	214.0	196.2	197.6	205.6
13	176.1	194.6	187.4	202.6	169.8	163.5	211.0	195.0	165.4	176.0
14			181.0	193.0			215.2	188.6		
15			187.2	199.8			223.2	207.0		
16			199.2	205.0			218.6	201.0		
17			197.4	206.4			220.2	198.0		
18			192.8	215.6			221.2	196.2		
19			191.0	213.4			214.8	196.6		
20			197.0	215.8			227.6	198.0		
21			205.6	210.8			234.0	192.6		
22			240.6	214.6			225.6	192.2		
23			222.2	224.8			213.8	190.6		
24			212.2	226.4			226.2	189.6		
25			217.2	235.4			235.6	192.0		
26			208.0	202.8			210.0	168.6		

APPENDIX 8

(Water consumption - continued)

Week	Group 4F: Air control				Group 5F: High dose	
	Cage					
	27	28	29	30	31	32
-1	177.0	188.4	163.0	171.4	160.4	176.4
1	180.4	163.0	165.4	163.6	157.6	177.4
2	173.4	157.4	159.6	161.8	155.8	176.6
3	174.0	158.6	157.0	157.6	162.0	174.2
4	175.6	167.6	163.6	158.0	169.8	238.6
5	188.0	161.0	161.6	165.0	155.6	286.6
6	185.8	167.4	172.0	165.6	160.8	278.4
7	185.4	173.6	178.8	168.2	163.0	(349.8)
8	180.6	169.0	164.2	161.2	167.2	265.2
9	183.6	177.8	174.2	176.2	179.6	301.4
10	193.2	184.6	173.2	178.6	(174.4)	324.2
11	202.4	178.0	188.8	176.8	(168.8)	373.0
12	197.0	175.0	182.2	179.6	(157.4)	401.8
13	177.0	163.8	169.8	154.8	(8.9)	292.6
14					196.0	205.2
15					189.8	240.2
16					184.0	303.0
17					181.8	256.4
18					180.3	283.2
19					168.5	283.0
20					172.0	267.0
21					169.0	292.6
22					153.0	243.8
23					146.3	241.6
24					151.0	214.2
25					154.0	221.0
26					145.5	226.0

( ) Value excluded from means and statistical analysis

APPENDIX 9

Ophthalmic examination - individual findings

Group	Animal number	Observations			
		Pre-dose		Week 13	
		Eye	Comments	Eye	Comments
1 M (Air control)	1		Nothing abnormal detected		Nothing abnormal detected
	2		NAD		NAD
	3		NAD		NAD
	4		NAD		NAD
	5		NAD		NAD
	6		NAD		NAD
	7		NAD		NAD
	8		NAD		NAD
	9		NAD		NAD
	10		NAD		NAD
	11		NAD		NAD
	12		NAD		NAD
	13		NAD		NAD
	14		NAD		NAD
	15		NAD		NAD
	16		NAD		NAD
	17		NAD		NAD
	18		NAD		NAD
	19		NAD		NAD
	20		NAD		NAD
2 M (NaF)	21		NAD		NAD
	22		NAD		NAD
	23		NAD		NAD
	24		NAD		NAD
	24		NAD		NAD
	26		NAD		NAD
	27		NAD		NAD
	28		NAD		NAD
	29		NAD		NAD
	30		NAD		NAD
	31		NAD		NAD
	32		NAD		NAD
	33		NAD		NAD
	34		NAD		NAD
	35		NAD		NAD
	36		NAD		NAD
	37		NAD		NAD
	38		NAD		NAD
	39		NAD		NAD
	40		NAD		NAD

APPENDIX 9

(Ophthalmic examination - individual findings - continued)

Group	Animal number	Observations			
		Pre-dose		Week 13	
		Eye	Comments	Eye	Comments
1 F (Air control)	81		Nothing abnormal detected		Nothing abnormal detected
	82		NAD		NAD
	83		NAD		NAD
	84		NAD		NAD
	85		NAD		NAD
	86		NAD		NAD
	87		NAD		NAD
	88		NAD		NAD
	89		NAD		NAD
	90		NAD		NAD
	91		NAD		NAD
	92		NAD		NAD
	93		NAD		NAD
	94		NAD		NAD
	95		NAD		NAD
	96		NAD		NAD
	97		NAD		NAD
	98		NAD		NAD
	99		NAD		NAD
	100		NAD		NAD
2 F (NaF)	101		NAD		NAD
	102		NAD		NAD
	103		NAD		NAD
	104		NAD		NAD
	105		NAD		NAD
	106		NAD		NAD
	107		NAD		NAD
	108		NAD		NAD
	109		NAD		NAD
	110		NAD		NAD
	111		NAD		NAD
	112		NAD		NAD
	113		NAD		NAD
	114		NAD		NAD
	115		NAD		NAD
	116		NAD		NAD
	117		NAD		NAD
	118		NAD		NAD
	119		NAD		NAD
	120		NAD		NAD

APPENDIX 9

(Ophthalmic examination - individual findings - continued)

Group	Animal number	Observations			
		Pre-dose		Week 13	
		Eye	Comments	Eye	Comments
3 M (Low dose Cryolite)	41		Nothing abnormal detected		
	42		NAD		
	43		NAD		
	44		NAD		
	45		NAD		
	46		NAD		
	47		NAD		
	48		NAD		
	49		NAD		
	50		NAD		
4 M (Inter dose Cryolite)	51		NAD		
	52		NAD		
	53		NAD		
	54		NAD		
	55		NAD		
	56		NAD		
	57		NAD		
	58		NAD		
	59		NAD		
	60		NAD		
5 M (High dose Cryolite)	61		NAD		NAD
	62		NAD		NAD
	63		NAD		NAD
	64		NAD		NAD
	65		NAD		NAD
	66		NAD		NAD
	67		NAD		NAD
	68		NAD		NAD
	69	R	Persistent pupillary membrane		NAD
		L	NAD		NAD
	70		NAD		NAD
	71		NAD		NAD
	72		NAD		NAD
	73		NAD		NAD
	74		NAD	R	Hyaloid remnants attached to posterior pole
				L	NAD
	75		NAD		NAD
	76		NAD		NAD
	77		NAD		NAD
	78		NAD		NAD
	79		NAD		NAD
	80		NAD		NAD

R Right eye
L Left eye

APPENDIX 9

(Ophthalmic examination - individual findings - continued)

Group	Animal number	Observations			
		Pre-dose		Week 13	
		Eye	Comments	Eye	Comments
3 F (Low dose Cryolite)	121		Nothing abnormal detected		
	122		NAD		
	123		NAD		
	124		NAD		
	125		NAD		
	126		NAD		
	127		NAD		
	128		NAD		
	129		NAD		
	130		NAD		
4 F (Inter dose Cryolite)	131		NAD		
	132		NAD		
	133		NAD		
	134		NAD		
	135		NAD		
	136		NAD		
	137		NAD		
	138		NAD		
	139		NAD		
	140		NAD		
5 F (High dose Cryolite)	141		NAD		NAD
	142		NAD		NAD
	143		NAD		NAD
	144		NAD		NAD
	145		NAD		NAD
	146		NAD		NAD
	147		NAD		NAD
	148		NAD		NAD
	149		NAD		NAD
	150		NAD		NAD
	151		NAD		NAD
	152		NAD		NAD
	153		NAD		NAD
	154		NAD		NAD
	155		NAD		NAD
	156		NAD		NAD
	157		NAD		NAD
	158		NAD		NAD
	159		NAD		NAD
	160		NAD		NAD

APPENDIX 10

Haematology - individual values

Week 13 (5 November 1996)

Group	Animal no.	PCV %	Hb g/dl	RBC $10^{12}/l$	MCHC g/dl	MCV fl	MCH pg	Retic %	WBC Total $10^9/l$	N $10^9/l$	L $10^9/l$	E $10^9/l$	B $10^9/l$
1M Air Control	1	44.3	16.2	8.88	36.5	49.9	18.2	1.0	11.29	1.89	9.12	0.09	0.01
	2	46.9	16.7	8.70	35.5	54.0	19.2	1.3	8.93	1.89	6.61	0.25	0.02
	3	43.8	15.5	8.31	35.4	52.7	18.7	1.6	13.99	5.29	7.66	0.51	0.04
	4	40.9	14.4	8.41	35.2	48.6	17.1	1.1	21.40	2.80	17.67	0.40	0.08
	5	46.3	16.3	8.95	35.1	51.7	18.2	1.3	6.39	1.11	5.09	0.12	0.01
	6	46.8	16.4	9.06	35.1	51.6	18.1	1.3	8.47	1.18	7.01	0.14	0.02
	7	43.8	15.5	8.23	35.3	53.2	18.8	2.3	8.39	1.26	6.66	0.29	0.01
	8	44.8	15.7	8.47	35.0	52.8	18.5	1.0	11.57	1.08	10.14	0.16	0.03
	9	47.0	16.4	8.56	35.0	54.9	19.2	1.6	9.32	0.97	8.02	0.18	0.02
	10	44.5	15.3	8.58	34.4	51.9	17.9	1.5	9.94	1.61	8.09	0.12	0.02
	Mean	44.9	15.8	8.62	35.3	52.1	18.4	1.4	10.97	1.91	8.61	0.23	0.03
	sd	1.91	0.69	0.278	0.53	1.86	0.64	0.39	4.219	1.311	3.479	0.138	0.021
2M NaF	21	44.2	15.6	8.71	35.3	50.8	17.9	1.5	9.62	3.17	5.79	0.28	0.02
	22	43.7	15.1	8.19	34.5	53.4	18.4	2.7	9.88	2.69	6.84	0.16	0.02
	23	42.0	14.8	8.23	35.3	51.0	18.0	1.2	12.34	2.14	6.99	0.21	0.04
	24	46.4	16.6	8.35	35.7	55.5	19.8	1.6	9.20	0.65	8.33	0.11	0.02
	25	42.9	14.8	7.94	34.5	54.0	18.6	4.0	18.49	2.14	15.73	0.23	0.06
	26	45.2	15.6	8.44	34.6	53.5	18.5	1.8	7.63	1.60	5.87	0.08	0.01
	27	44.5	15.4	8.78	34.7	50.6	17.6	1.6	9.97	1.12	8.59	0.11	0.02
	28	43.9	15.3	8.57	34.9	51.3	17.9	1.5	13.97	1.97	11.49	0.22	0.04
	29	44.9	15.9	8.72	35.4	51.5	18.2	3.0	11.88	1.44	10.10	0.15	0.03
	30	43.5	14.9	8.31	34.3	52.3	18.0	1.6	14.87	1.82	12.53	0.18	0.04
	Mean	44.1	15.4	8.42	34.9	52.4	18.3	2.1	11.79	1.87	9.23	0.17	0.03
	sd	1.23	0.56	0.271	0.47	1.64	0.61	0.89	3.251	0.733	3.233	0.063	0.015

sd Standard deviation

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	M $10^9/l$	LUC $10^9/l$	Anis	Micro	Macro	Var	Hypo	Hyper	LS	Atyp	Blast	Plt $10^9/l$
1M Air Control	1	0.11	0.07	-	+	-	+	-	+	-	-	-	1001
	2	0.04	0.12	-	-	-	+	-	+	-	-	-	888
	3	0.23	0.26	-	-	-	+	-	+	-	-	-	971
	4	0.25	0.21	-	+	-	+	-	+	-	-	-	1344
	5	0.03	0.03	-	-	-	+	-	-	-	-	-	967
	6	0.08	0.05	-	-	-	+	-	-	-	-	-	877
	7	0.08	0.09	-	-	-	+	-	-	-	-	-	924
	8	0.10	0.07	-	-	-	-	-	-	-	-	-	1005
	9	0.07	0.05	-	-	-	-	-	-	-	-	-	952
	10	0.07	0.03	-	-	-	+	-	-	-	-	-	795
	Mean	0.11	0.10										972
	sd	0.075	0.078										145.5
2M NaF	21	0.24	0.11	-	-	-	+	-	-	-	-	-	880
	22	0.09	0.07	-	-	-	+	-	-	-	-	-	900
	23	2.88	0.08	-	-	-	+	-	-	-	-	-	1205
	24	0.06	0.03	-	-	-	+	-	+	-	-	-	1039
	25	0.15	0.19	-	-	-	++	-	+	-	-	-	985
	26	0.05	0.01	-	-	-	+	-	-	-	-	-	938
	27	0.08	0.06	-	-	-	-	-	-	-	-	-	840
	28	0.15	0.10	-	-	-	+	-	-	-	-	-	1055
	29	0.09	0.08	-	-	-	+	-	+	-	-	-	908
	30	0.14	0.15	-	-	-	+	-	-	-	-	-	940
	Mean	0.39	0.09										969
	sd	0.876	0.053										107.0

sd Standard deviation

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	PCV %	Hb g/dl	RBC $10^{12}/l$	MCHC g/dl	MCV fl	MCH pg	Retic %	WBC Total $10^9/l$	N $10^9/l$	L $10^9/l$	E $10^9/l$	B $10^9/l$
3M Low dose	41	42.9	15.3	8.40	35.6	51.1	18.2	0.8	9.69	1.52	7.87	0.11	0.02
	42	45.8	16.1	8.88	35.2	51.6	18.2	1.3	9.75	1.79	7.69	0.08	0.03
	43	44.6	15.6	8.49	35.0	52.5	18.4	1.7	8.10	1.52	6.40	0.09	0.02
	44	44.9	15.7	8.59	35.0	52.3	18.3	3.2	10.46	2.63	7.56	0.12	0.02
	45	45.5	15.6	9.09	34.3	50.1	17.2	2.3	10.99	2.45	8.05	0.19	0.02
	46	41.9	14.8	8.11	35.4	51.6	18.3	1.3	13.79	1.79	11.73	0.11	0.03
	47	45.8	15.9	8.42	34.7	54.4	18.9	1.2	6.63	0.70	5.72	0.12	0.01
	48	46.5	16.1	8.61	34.6	54.0	18.7	2.0	6.76	1.29	5.28	0.10	0.02
	49	47.5	16.6	9.12	35.0	52.1	18.2	1.6	9.83	2.62	6.79	0.26	0.03
	50	48.1	16.9	9.07	35.2	53.1	18.7	1.0	6.94	1.16	5.53	0.15	0.01
	Mean	45.4	15.9	8.68	35.0	52.3	18.3	1.6	9.29	1.75	7.26	0.13	0.02
	sd	1.90	0.61	0.345	0.39	1.30	0.46	0.71	2.251	0.649	1.871	0.055	0.007
4M Inter dose	51	45.4	16.0	8.66	35.3	52.4	18.5	2.1	10.10	4.67	5.07	0.18	0.02
	52	47.9	16.7	8.94	34.9	53.6	18.7	1.7	12.82	1.86	10.60	0.15	0.03
	53	47.8	16.6	9.04	34.7	52.9	18.4	2.0	16.23	1.99	13.75	0.26	0.06
	54	44.2	15.2	8.02	34.5	55.1	19.0	1.8	15.16	5.35	8.95	0.53	0.04
	55	44.7	15.1	8.81	33.8	50.8	17.2	3.7	11.55	2.50	8.49	0.23	0.02
	56	47.1	16.2	8.78	34.4	53.6	18.4	3.0	9.54	1.59	7.71	0.09	0.02
	57	44.9	15.5	8.28	34.5	54.3	18.7	2.8	12.64	1.76	10.62	0.11	0.03
	58	44.7	15.2	8.34	34.1	53.6	18.3	2.2	13.48	2.37	10.77	0.18	0.04
	59	44.1	15.6	8.62	35.3	51.2	18.1	2.4	11.71	2.81	8.32	0.37	0.02
	60	44.7	15.7	8.54	35.1	52.3	18.4	2.3	12.23	1.72	9.87	0.27	0.04
	Mean	45.6	15.8	8.60	34.7	53.0	18.4	2.4	12.55	2.66	9.42	0.24	0.03
	sd	1.47	0.58	0.316	0.50	1.34	0.48	0.61	2.059	1.305	2.303	0.132	0.013

sd Standard deviation

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	M $10^9/l$	LUC $10^9/l$	Anis	Micro	Macro	Var	Hypo	Hyper	LS	Atyp	Blast	Plt $10^9/l$
3M Low dose	41	0.08	0.10	-	-	-	+	-	-	-	-	-	997
	42	0.07	0.09	-	-	-	+	-	+	-	-	-	728
	43	0.05	0.02	-	-	-	+	-	+	-	-	-	924
	44	0.07	0.06	-	-	-	++	-	+	-	-	-	1052
	45	0.18	0.10	-	-	-	+	-	-	-	-	-	876
	46	0.07	0.05	-	-	-	+	-	+	-	-	-	1046
	47	0.04	0.03	-	-	-	+	-	-	-	-	-	922
	48	0.04	0.03	-	-	-	+	-	+	-	-	-	919
	49	0.09	0.04	-	-	-	+	-	-	-	-	-	926
	50	0.06	0.04	-	-	-	+	-	+	-	-	-	915
	Mean	0.08	0.06										931
	sd	0.040	0.030										92.5
4M Inter dose	51	0.08	0.09	-	-	-	+	-	-	-	-	-	848
	52	0.12	0.07	-	-	-	+	-	-	-	-	-	995
	53	0.11	0.07	-	-	-	+	-	+	-	-	-	1176
	54	0.19	0.10	-	-	-	+	-	+	-	-	-	929
	55	0.18	0.14	-	+	-	++	-	-	-	-	-	949
	56	0.09	0.04	-	-	-	-	-	-	-	-	-	872
	57	0.07	0.05	-	-	-	+	-	+	-	-	-	913
	58	0.08	0.06	-	-	-	+	-	-	-	-	-	1072
	59	0.19	0.00	-	-	-	+	-	-	-	-	-	985
	60	0.13	0.19	-	-	-	+	-	-	-	-	-	879
	Mean	0.12	0.08										962
	sd	0.047	0.053										100.6

sd Standard deviation

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	PCV %	Hb g/dl	RBC $10^{12}/l$	MCHC g/dl	MCV fl	MCH pg	Retic %	WBC Total $10^9/l$	N $10^9/l$	L $10^9/l$	E $10^9/l$	B $10^9/l$
5M High dose	61	43.9	15.3	8.64	34.8	50.9	17.7	2.6	17.19	2.53	13.87	0.34	0.06
	62	44.6	15.5	9.03	34.8	49.4	17.2	0.9	7.18	1.81	5.04	0.23	0.01
	63	46.6	16.3	9.01	35.1	51.7	18.1	3.2	12.75	1.37	11.10	0.12	0.04
	64	46.5	16.4	8.96	35.2	51.8	18.3	3.3	12.19	2.48	9.30	0.15	0.04
	65	46.4	16.4	8.79	35.3	52.8	18.6	1.3	12.92	1.78	10.70	0.25	0.02
	66	45.1	15.5	8.75	34.4	51.6	17.8	1.6	12.69	2.60	9.44	0.24	0.02
	67	43.5	15.2	8.31	34.9	52.4	18.3	1.6	13.81	1.39	11.26	0.93	0.05
	68	45.5	16.1	9.14	35.4	49.8	17.6	2.1	13.90	2.22	11.26	0.16	0.05
	69	44.3	15.1	8.45	34.2	52.4	17.9	2.2	16.41	3.93	11.86	0.24	0.05
	70	44.6	16.2	8.89	36.3	50.2	18.2	2.7	25.53	1.77	22.76	0.34	0.12
	Mean	45.1	15.8	8.80	35.0	51.3	18.0	2.2	14.46	2.19	11.66	0.30	0.05
	sd	1.12	0.53	0.266	0.58	1.17	0.41	0.80	4.727	0.760	4.522	0.233	0.031

sd Standard deviation

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	M $10^9/l$	LUC $10^9/l$	Anis	Micro	Macro	Var	Hypo	Hyper	LS	Atyp	Blast	Plt $10^9/l$
5M High dose	61	0.23	0.16	-	-	-	+	-	-	-	-	-	736
	62	0.07	0.02	-	-	-	+	-	-	-	-	-	885
	63	0.09	0.04	-	-	-	+	-	-	-	-	-	938
	64	0.14	0.08	-	-	-	+	-	-	-	-	-	1010
	65	0.11	0.05	-	-	-	-	-	-	-	-	-	1071
	66	0.26	0.14	-	-	-	+	-	-	-	-	-	1062
	67	0.10	0.08	-	-	-	+	-	-	-	-	-	780
	68	0.12	0.07	-	-	-	+	-	-	-	-	-	848
	69	0.19	0.13	-	-	-	+	-	-	-	-	-	819
	70	0.18	0.35	-	+	-	+	-	-	-	-	-	879
	Mean	0.15	0.11										903
	sd	0.063	0.095										115.6

sd Standard deviation

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	PCV %	Hb g/dl	RBC $10^{12}/l$	MCHC g/dl	MCV fl	MCH pg	Retic %	WBC Total $10^9/l$	N $10^9/l$	L $10^9/l$	E $10^9/l$	B $10^9/l$
1F Air Control	81	43.0	15.1	7.67	35.2	56.1	19.8	1.4	7.79	0.79	6.77	0.09	0.01
	82	42.1	15.2	7.83	36.2	53.7	19.5	0.9	5.03	0.64	4.05	0.16	0.01
	83	41.3	14.5	7.23	35.1	57.1	20.1	1.4	8.37	0.72	7.29	0.19	0.01
	85	39.3	14.3	7.64	36.3	51.5	18.7	0.5	13.63	1.23	12.22	0.08	0.04
	86	44.1	15.9	7.86	36.0	56.1	20.2	1.4	5.98	0.75	5.02	0.14	0.01
	87	42.4	15.2	7.54	35.9	56.3	20.2	1.0	6.97	1.52	5.02	0.22	0.02
	88	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd
	89	43.8	15.9	7.88	36.2	55.6	20.1	1.7	7.45	1.02	6.16	0.17	0.02
	90	41.0	14.7	7.49	35.9	54.8	19.7	2.3	8.02	1.22	6.58	0.08	0.02
	Mean	42.1	15.1	7.64	35.9	55.2	19.8	1.3	7.91	0.99	6.64	0.14	0.02
	sd	1.58	0.59	0.222	0.46	1.80	0.51	0.54	2.565	0.313	2.500	0.053	0.010
2F NaF	101	41.2	14.9	7.98	36.1	51.6	18.7	0.9	7.13	0.65	6.25	0.10	0.01
	102	45.4	16.1	8.25	35.6	55.0	19.6	2.3	5.24	0.98	4.12	0.08	0.01
	103	42.6	15.1	7.88	35.5	54.1	19.2	1.5	9.69	0.91	8.53	0.15	0.02
	104	42.6	15.1	8.04	35.5	53.0	18.8	1.8	6.52	0.75	5.55	0.12	0.01
	105	42.7	15.2	7.45	35.5	57.3	20.4	1.6	14.02	0.86	12.88	0.14	0.05
	107	39.8	14.0	7.60	35.2	52.3	18.4	1.3	6.78	0.50	6.08	0.11	0.01
	108	43.1	15.4	7.88	35.6	54.7	19.5	1.6	10.05	1.04	8.68	0.17	0.02
	109	45.3	15.7	8.07	34.7	56.1	19.5	1.2	9.53	1.03	8.19	0.10	0.02
	110	43.1	15.3	7.90	35.4	54.6	19.3	2.2	12.61	0.86	11.38	0.13	0.04
	Mean	42.9	15.2	7.89	35.5	54.3	19.3	1.6	9.06	0.84	7.96	0.12	0.02
	sd	1.76	0.58	0.242	0.37	1.80	0.59	0.45	2.932	0.181	2.824	0.028	0.015
3F Low dose	121	43.1	15.1	7.71	35.0	55.9	19.6	1.7	7.87	0.92	6.67	0.10	0.02
	122	43.6	15.4	7.89	35.3	55.3	19.5	2.6	6.72	0.86	5.43	0.10	0.01
	123	43.0	15.4	7.64	35.9	56.4	20.2	1.2	9.17	1.87	6.72	0.30	0.01
	124	41.2	14.5	7.52	35.2	54.8	19.3	1.0	9.02	1.07	7.60	0.15	0.02
	125	41.9	14.9	7.35	35.5	57.0	20.2	1.6	7.24	1.08	5.85	0.20	0.01
	126	41.8	14.9	7.29	35.6	57.3	20.4	2.2	6.18	1.09	4.89	0.11	0.01
	127	43.1	15.0	7.77	34.8	55.4	19.3	1.8	7.59	1.45	5.55	0.28	0.02
	128	42.2	14.8	7.88	34.9	53.6	18.7	2.4	5.77	0.79	4.76	0.14	0.01
	129	42.2	14.9	7.55	35.4	55.9	19.8	1.3	6.23	1.14	4.88	0.12	0.01
	130	43.9	15.3	7.99	34.9	54.9	19.2	3.7	5.45	0.61	4.63	0.11	0.02
	Mean	42.6	15.0	7.66	35.3	55.7	19.6	2.0	7.12	1.09	5.70	0.16	0.01
	sd	0.87	0.29	0.233	0.36	1.10	0.53	0.81	1.293	0.356	1.004	0.074	0.005

sd Standard deviation
ctd Clotted Sample

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	M $10^9/l$	LUC $10^9/l$	Anis	Micro	Macro	Var	Hypo	Hyper	LS	Atyp	Blast	Plt $10^9/l$
1F Air Control	81	0.08	0.06	-	-	-	-	-	-	-	-	-	1221
	82	0.10	0.08	-	-	-	-	-	-	-	-	-	960
	83	0.07	0.08	-	-	-	-	-	-	-	-	-	990
	85	0.05	0.02	-	-	-	+	-	-	-	-	-	1199
	86	0.04	0.02	-	-	-	-	-	-	-	-	-	926
	87	0.11	0.07	-	-	-	-	-	-	-	-	-	1003
	88	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd
	89	0.06	0.01	-	-	-	-	-	-	-	-	-	1010
	90	0.08	0.04	-	-	-	-	-	-	-	-	-	1038
	Mean	0.07	0.05										1043
	sd	0.024	0.029										108.3
2F NaF	101	0.08	0.05	-	-	-	-	-	-	-	-	-	973
	102	0.03	0.02	-	-	-	-	-	-	-	-	-	825
	103	0.04	0.03	-	-	-	-	-	-	-	-	-	978
	104	0.06	0.03	-	-	-	-	-	-	-	-	-	1022
	105	0.06	0.04	-	-	-	-	-	-	-	-	-	1194
	107	0.05	0.04	-	-	-	-	-	-	-	-	-	1024
	108	0.08	0.06	-	-	-	-	-	-	-	-	-	840
	109	0.12	0.07	-	-	-	-	-	-	-	-	-	866
	110	0.10	0.11	-	-	-	-	-	-	-	-	-	1018
	Mean	0.07	0.05										971
	sd	0.029	0.027										115.6
3F Low dose	121	0.09	0.07	-	-	-	-	-	-	-	-	-	859
	122	0.12	0.21	-	-	-	-	-	-	-	-	-	837
	123	0.12	0.14	-	-	-	-	-	-	-	-	-	1117
	124	0.09	0.08	-	-	-	-	-	-	-	-	-	1011
	125	0.05	0.05	-	-	-	-	-	-	-	-	-	1310
	126	0.05	0.03	-	-	-	-	-	-	-	-	-	1047
	127	0.14	0.15	-	-	-	-	-	-	-	-	-	864
	128	0.04	0.03	-	-	-	-	-	-	-	-	-	979
	129	0.05	0.04	-	-	-	-	-	-	-	-	-	1074
	130	0.04	0.05	-	-	-	-	-	-	-	-	-	840
	Mean	0.08	0.09										994
	sd	0.038	0.061										152.1

sd Standard deviation
ctd Clotted Sample

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	PCV %	Hb g/dl	RBC $10^{12}/l$	MCHC g/dl	MCV fl	MCH pg	Retic %	WBC Total $10^9/l$	N $10^9/l$	L $10^9/l$	E $10^9/l$	B $10^9/l$
4F Inter dose	131	42.8	15.2	7.67	35.5	55.8	19.8	1.8	11.64	1.45	9.76	0.19	0.03
	132	43.5	15.2	7.81	34.9	55.7	19.4	2.6	7.48	1.54	5.49	0.32	0.01
	133	42.7	14.7	7.76	34.5	55.0	19.0	2.0	10.70	1.43	8.77	0.23	0.02
	134	43.5	15.6	8.23	35.8	52.8	18.9	1.9	12.44	1.45	10.62	0.18	0.03
	135	41.9	14.8	7.91	35.3	53.0	18.7	1.6	11.84	1.75	9.56	0.17	0.02
	136	42.1	15.0	7.47	35.7	56.3	20.1	1.0	9.46	0.57	8.72	0.10	0.02
	137	42.8	15.0	7.73	35.2	55.4	19.5	1.9	10.38	1.76	8.03	0.30	0.03
	138	43.4	15.2	8.07	35.1	53.8	18.9	2.9	10.12	1.28	8.57	0.15	0.02
	139	44.7	15.7	8.34	35.2	53.6	18.9	1.3	8.05	0.93	6.87	0.11	0.02
	140	42.7	15.1	7.48	35.3	57.1	20.2	2.3	11.22	1.59	9.29	0.17	0.02
	Mean	43.0	15.2	7.85	35.3	54.9	19.3	1.9	10.33	1.38	8.57	0.19	0.02
	sd	0.80	0.31	0.293	0.38	1.47	0.54	0.57	1.616	0.370	1.485	0.073	0.006
5F High dose	141	43.2	15.0	8.16	34.7	53.0	18.4	1.3	8.95	1.17	7.28	0.16	0.03
	142	43.0	15.1	8.01	35.2	53.7	18.9	0.9	8.43	1.35	6.60	0.15	0.03
	143	43.1	15.3	7.61	35.4	56.6	20.1	1.7	12.43	1.48	10.56	0.16	0.03
	144	42.4	14.9	7.43	35.2	57.1	20.1	2.1	7.28	1.20	5.65	0.16	0.02
	145	44.1	15.7	8.03	35.6	54.9	19.6	1.6	23.67	1.97	20.83	0.30	0.12
	146	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd
	147	41.5	14.5	7.56	35.1	54.8	19.2	1.5	9.37	1.17	7.85	0.14	0.03
	148	42.0	15.0	7.84	35.8	53.5	19.2	1.4	12.55	1.79	10.17	0.30	0.04
	149	46.0	16.0	8.28	34.8	55.5	19.3	2.0	14.77	2.44	11.94	0.14	0.03
	150	43.6	15.3	7.65	35.2	57.1	20.1	1.6	10.95	1.36	9.22	0.16	0.02
	Mean	43.2	15.2	7.84	35.2	55.1	19.4	1.6	12.04	1.55	10.01	0.19	0.04
	sd	1.31	0.44	0.295	0.35	1.56	0.60	0.36	4.958	0.436	4.535	0.065	0.031

sd Standard deviation
ctd Clotted Sample

APPENDIX 10

(Haematology - continued)

Week 13 (5 November 1996)

Group	Animal no.	M $10^9/l$	LUC $10^9/l$	Anis	Micro	Macro	Var	Hypo	Hyper	LS	Atyp	Blast	Plt $10^9/l$
4F Inter dose	131	0.11	0.10	-	-	-	-	-	-	-	-	-	958
	132	0.06	0.07	-	-	-	-	-	-	-	-	-	1069
	133	0.14	0.11	-	-	-	-	-	-	-	-	-	892
	134	0.07	0.10	-	-	-	-	-	-	-	-	-	933
	135	0.19	0.14	-	-	-	-	-	-	-	-	-	982
	136	0.03	0.01	-	-	-	+	-	-	-	-	-	876
	137	0.11	0.15	-	-	-	-	-	-	-	-	-	763
	138	0.05	0.04	-	-	-	-	-	-	-	-	-	919
	139	0.07	0.05	-	-	-	-	-	-	-	-	-	956
	140	0.09	0.06	-	-	-	-	-	-	-	-	-	1012
	Mean	0.09	0.08										936
	sd	0.047	0.045										83.2
5F High dose	141	0.17	0.14	-	-	-	-	-	-	-	-	-	912
	142	0.13	0.17	-	-	-	-	-	-	-	-	-	1102
	143	0.08	0.12	-	-	-	-	-	-	-	-	-	1144
	144	0.09	0.16	-	-	-	-	-	-	-	-	-	825
	145	0.23	0.21	-	-	-	-	-	-	-	-	-	693
	146	0.10	0.09	-	-	-	-	-	-	-	-	-	901
	147	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd	ctd
	148	0.13	0.13	-	-	-	-	-	-	-	-	-	809
	149	0.12	0.10	-	-	-	-	-	-	-	-	-	981
	150	0.12	0.06	-	-	-	-	-	-	-	-	-	1041
	Mean	0.13	0.13										934
	sd	0.046	0.045										147.1

sd Standard deviation
ctd Clotted Sample

APPENDIX 11

Biochemistry - individual values

Week 13 (5 November 1996)

Group	Animal no.	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	γGT mU/ml	gGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
1M Air Control	1	113	7.2	3.0	4.2	14	0.5	162	31	72	1	64	0.1	146	3.4	5.5	3.8	103	75
	2	87	7.1	3.1	4.0	16	0.6	144	32	72	<1	220	0.2	147	3.4	5.6	3.8	103	89
	3	125	6.5	2.8	3.7	16	0.5	224	24	73	<1	75	0.2	145	3.3	5.4	3.7	102	67
	4	118	5.9	2.9	3.0	17	0.7	141	37	84	<1	139	0.2	145	3.6	5.3	4.3	103	49
	5	81	6.9	3.0	3.9	18	0.6	246	38	78	<1	269	0.2	146	3.0	5.4	3.7	102	56
	6	110	6.8	2.9	3.9	16	0.6	133	31	63	<1	56	0.1	146	3.7	5.3	3.8	104	50
	7	83	7.0	3.0	4.0	16	0.5	230	36	68	<1	86	0.2	147	3.0	5.5	3.3	105	53
	8	121	6.9	2.8	4.1	16	0.6	159	29	56	<1	65	0.3	146	3.5	5.5	3.5	104	70
	9	111	6.8	2.9	3.9	18	0.5	167	27	55	2	58	0.1	145	3.2	5.4	3.3	103	55
	10	111	6.7	2.9	3.8	15	0.6	187	33	63	<1	55	0.2	147	3.4	5.4	3.6	105	57
	Mean	106	6.8	2.9	3.9	16	0.6	179	32	68	<1	109	0.2	146	3.4	5.4	3.7	103	62
	sd	16.2	0.37	0.09	0.33	1.2	0.07	40.5	4.4	9.3		76.6	0.06	0.8	0.23	0.09	0.29	1.1	12.9
2M NaF	21	99	7.1	3.0	4.1	16	0.6	150	31	68	1	144	0.2	146	3.4	5.4	4.0	102	44
	22	115	6.4	2.9	3.5	15	0.5	237	39	75	<1	101	0.1	147	4.2	5.2	3.7	107	56
	23	90	6.9	2.2	4.7	17	0.6	204	29	56	3	67	0.1	147	3.3	5.4	3.9	104	35
	24	101	6.9	2.8	4.1	16	0.6	157	33	93	<1	81	0.1	147	3.3	5.7	3.6	103	63
	25	152	6.8	2.8	4.0	12	0.6	176	28	83	<1	75	0.2	143	3.1	5.4	3.6	100	57
	26	109	6.4	2.7	3.7	15	0.6	163	33	67	1	54	0.1	146	3.7	5.4	3.6	105	71
	27	93	6.4	2.7	3.7	14	0.6	241	31	68	<1	112	0.2	146	3.3	5.4	3.5	103	53
	28	123	6.8	2.9	3.9	14	0.7	163	33	74	<1	79	0.2	145	3.4	5.4	3.7	103	46
	29	100	6.6	2.9	3.7	16	0.5	177	31	76	<1	48	0.1	146	3.4	5.1	3.4	103	57
	30	105	7.1	2.9	4.2	18	0.7	191	30	86	<1	216	0.1	147	3.8	5.4	3.5	105	63
	Mean	109	6.7	2.8	4.0	15	0.6	186	32	75	<1	98	0.1	146	3.5	5.4	3.7	104	55
	sd	18.1	0.28	0.23	0.34	1.7	0.07	32.2	3.0	10.7		50.4	0.05	1.2	0.32	0.15	0.18	1.9	10.5

sd Standard deviation

APPENDIX 11

(Biochemistry - continued)

Week 13 (5 November 1996)

Group	Animal no.	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	GOT mU/ml	γGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
3M Low dose	41	111	6.6	2.9	3.7	16	0.6	166	33	87	1	301	0.2	146	4.0	5.3	3.6	105	55
	42	125	7.1	3.0	4.1	17	0.6	183	23	67	<1	146	0.2	147	3.3	5.5	3.5	105	48
	43	124	6.5	2.9	3.6	17	0.5	148	30	74	<1	59	0.1	146	3.5	5.5	4.1	104	45
	44	128	7.2	2.7	4.5	18	0.6	178	24	61	<1	102	0.2	147	3.8	5.6	3.6	104	91
	45	129	6.7	2.9	3.8	19	0.6	241	36	79	<1	88	0.1	147	3.3	5.4	4.4	103	61
	46	127	6.5	3.0	3.5	17	0.6	213	30	61	<1	63	0.1	146	3.5	5.3	3.9	104	50
	47	100	6.0	2.5	3.5	16	0.5	185	30	85	1	86	0.2	147	3.6	5.1	3.9	106	43
	48	106	6.6	2.8	3.8	17	0.6	153	29	66	<1	61	0.1	147	3.6	5.4	3.7	105	50
	49	118	7.1	2.8	4.3	15	0.6	164	41	61	<1	48	0.2	146	3.7	5.5	3.3	104	56
	50	98	7.1	2.9	4.2	17	0.6	197	32	79	<1	65	0.2	148	3.7	5.6	3.9	104	47
	Mean	117	6.7	2.8	3.9	17	0.6	183	31	72	<1	102	0.2	147	3.6	5.4	3.8	104	55
	sd	11.9	0.38	0.15	0.35	1.1	0.04	28.4	5.3	10.1		75.5	0.05	0.7	0.22	0.15	0.32	0.8	13.9
4M Inter dose	51	146	7.2	3.0	4.2	14	0.5	228	31	78	1	56	0.1	146	3.5	5.6	3.6	103	60
	52	127	6.8	2.9	3.9	18	0.5	136	33	58	<1	116	0.1	147	3.7	5.4	3.7	104	59
	53	124	6.7	2.7	4.0	15	0.5	156	26	69	<1	353	0.2	147	4.1	5.5	4.0	104	67
	54	122	6.8	2.9	3.9	16	0.5	125	27	57	<1	60	0.2	148	3.8	5.7	4.3	105	69
	55	119	7.2	3.0	4.2	16	0.5	187	29	58	<1	90	0.2	146	3.8	5.7	4.1	103	81
	56	129	7.0	3.0	4.0	17	0.5	194	31	72	<1	73	0.1	146	3.9	5.6	4.0	103	58
	57	112	6.7	2.9	3.8	14	0.5	235	24	67	<1	54	0.1	147	3.9	5.5	4.4	104	59
	58	111	6.3	2.8	3.5	17	0.6	172	28	66	<1	60	0.1	147	3.5	5.5	4.0	105	50
	59	121	6.7	2.9	3.8	18	0.6	155	25	67	<1	254	0.2	145	4.0	5.3	3.8	104	59
	60	105	6.2	2.8	3.4	13	0.5	172	28	59	1	58	0.1	146	4.0	5.4	4.0	104	62
	Mean	122	6.8	2.9	3.9	16	0.5	176	28	65	<1	117	0.1	147	3.8	5.5	4.0	104	62
	sd	11.4	0.33	0.10	0.26	1.8	0.04	36.1	2.9	7.0		102.6	0.05	0.8	0.20	0.13	0.25	0.7	8.3

sd Standard deviation

APPENDIX 11

(Biochemistry - continued)

Week 13 (5 November 1996)

Group	Animal no.	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	GOT mU/ml	γGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
5M High dose	61	110	6.7	2.8	3.9	17	0.5	178	31	69	<1	129	0.2	146	3.9	5.6	4.7	103	61
	62	120	6.5	2.8	3.7	14	0.5	200	31	87	1	76	0.2	148	3.9	5.4	4.4	106	51
	63	120	6.8	3.0	3.8	13	0.5	193	31	67	1	75	0.2	148	3.4	5.4	4.1	105	48
	64	112	6.8	3.0	3.8	13	0.5	203	33	67	<1	63	0.2	147	3.8	5.5	4.1	102	54
	65	125	7.1	3.1	4.0	17	0.6	197	28	65	<1	97	0.2	146	3.5	5.5	3.7	103	49
	66	101	6.9	3.0	3.9	17	0.5	196	23	53	<1	249	0.2	146	3.7	5.8	4.0	103	74
	67	120	6.5	2.8	3.7	15	0.5	178	37	75	<1	78	0.2	146	3.5	5.5	4.5	103	68
	68	114	7.2	2.9	4.3	15	0.5	190	27	69	1	150	0.1	147	3.8	5.7	4.1	105	84
	69	115	6.6	2.8	3.8	20	0.6	163	28	72	<1	56	<0.1	146	3.9	5.4	4.2	104	57
	70	138	6.6	3.0	3.6	19	0.6	198	28	67	<1	94	0.1	146	3.1	5.4	4.5	104	70
	Mean	118	6.8	2.9	3.9	16	0.5	190	30	69	<1	107	<0.2	147	3.7	5.5	4.2	104	62
	sd	9.8	0.24	0.11	0.20	2.4	0.05	12.7	3.8	8.5		57.9		0.8	0.27	0.14	0.29	1.2	12.0

sd Standard deviation

APPENDIX 11

(Biochemistry - continued)

Week 13 (5 November 1996)

Group	Animal no.	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	GOT mU/ml	γGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
1F Air Control	81	106	6.9	3.3	3.6	24	0.7	78	21	56	1	110	0.2	144	3.9	5.4	3.3	104	79
	82	96	7.6	3.4	4.2	20	0.7	85	36	76	<1	55	0.2	145	3.4	5.5	3.3	103	89
	83	91	7.1	3.4	3.7	19	0.6	77	34	74	1	97	0.2	147	3.1	5.5	3.3	104	73
	85	125	7.3	2.9	4.4	18	0.5	130	30	72	<1	52	0.2	144	2.9	5.4	3.4	101	58
	86	109	7.3	3.4	3.9	20	0.7	201	30	73	1	89	0.2	147	3.1	5.4	2.7	105	50
	87	102	6.6	3.1	3.5	26	0.7	75	28	63	<1	160	0.2	145	3.1	5.3	3.5	105	79
	88	145	7.3	3.2	4.1	20	0.7	111	19	46	<1	36	0.3	144	2.9	5.3	2.7	102	63
	89	104	7.2	3.0	4.2	21	0.7	111	23	59	<1	62	0.2	145	3.2	5.5	3.7	104	87
	90	134	7.3	3.3	4.0	18	0.7	90	25	72	<1	60	0.2	145	2.7	5.4	2.4	103	99
	Mean	112	7.2	3.2	4.0	21	0.7	106	27	66	<1	80	0.2	145	3.1	5.4	3.1	103	75
	sd	18.2	0.29	0.19	0.30	2.7	0.07	40.2	5.8	10.3		38.3	0.03	1.2	0.35	0.08	0.44	1.3	15.8
2F NaF	101	123	7.2	3.4	3.8	18	0.6	89	29	58	<1	58	0.2	145	3.6	5.6	3.6	104	68
	102	131	7.3	3.3	4.0	23	0.7	96	24	69	<1	201	0.3	146	3.1	5.4	2.7	105	83
	103	105	7.3	3.4	3.9	19	0.7	97	36	71	<1	47	0.2	144	3.6	5.4	3.3	103	57
	104	109	7.4	3.4	4.0	17	0.6	87	23	51	<1	49	0.2	146	3.6	5.5	3.5	105	80
	105	88	6.7	2.9	3.8	17	0.6	92	19	58	1	53	0.2	144	3.5	5.3	3.5	104	38
	107	133	8.0	3.9	4.1	19	0.6	62	33	75	<1	44	0.2	146	3.3	5.6	3.5	103	75
	108	139	6.9	2.9	4.0	22	0.7	118	31	88	<1	84	0.2	144	3.6	5.5	4.1	103	79
	109	128	7.1	3.1	4.0	17	0.6	92	33	65	<1	62	0.1	148	3.8	5.5	3.9	107	79
	110	144	7.7	3.6	4.1	20	0.6	68	27	56	<1	133	0.2	145	3.3	5.8	3.8	103	70
	Mean	122	7.3	3.3	4.0	19	0.6	89	28	66	<1	81	0.2	145	3.5	5.5	3.5	104	70
	sd	18.1	0.39	0.32	0.11	2.2	0.05	16.4	5.5	11.5		52.8	0.05	1.3	0.21	0.15	0.40	1.4	14.4
3F Low dose	121	117	7.9	3.8	4.1	18	0.7	98	44	101	<1	67	0.3	144	3.3	5.8	3.3	101	77
	122	121	7.4	3.5	3.9	23	0.7	89	37	101	1	880	0.2	144	3.9	5.6	3.2	103	81
	123	107	6.8	2.9	3.9	22	0.7	117	28	66	<1	108	0.2	147	3.8	5.3	3.2	106	56
	124	120	7.4	3.2	4.2	27	0.8	95	51	79	<1	106	0.1	145	3.2	5.6	3.0	104	64
	125	156	7.6	3.4	4.2	17	0.6	92	29	67	<1	48	0.2	145	3.6	5.5	2.3	105	88
	126	109	7.7	3.4	4.3	16	0.6	96	28	92	1	279	0.1	146	3.1	5.6	2.5	103	94
	127	94	6.9	2.8	4.1	23	0.7	111	22	65	1	172	0.1	146	3.7	5.4	3.7	105	74
	128	130	6.7	3.3	3.4	24	0.8	85	33	63	<1	55	0.2	146	3.4	5.4	2.7	106	48
	129	93	7.0	3.3	3.7	28	0.9	107	25	66	<1	247	0.1	147	3.4	5.5	3.3	105	62
	130	132	7.4	3.4	4.0	16	0.6	109	39	73	<1	60	0.2	147	3.2	5.5	2.8	105	77
	Mean	118	7.3	3.3	4.0	21	0.7	100	34	77	<1	202	0.2	146	3.5	5.5	3.0	104	72
	sd	18.9	0.41	0.29	0.27	4.4	0.10	10.5	9.1	15.2		251.7	0.07	1.2	0.28	0.14	0.42	1.6	14.4

sd Standard deviation

APPENDIX 11

(Biochemistry - continued)

Week 13 (5 November 1996)

Group	Animal no.	Glu-cose mg/dl	Protein g/dl Total	Alb	Glob	Urea Nitr mg/dl	Creat-inine mg/dl	AP mU/ml	GPT mU/ml	GOT mU/ml	γGT mU/ml	CPK mU/ml	Bili-rubin mg/dl	Na mEq/l	K mEq/l	Ca mEq/l	P mEq/l	Cl mEq/l	Chol mg/dl
4F Inter dose	131	96	7.2	3.4	3.8	23	0.7	104	84	118	2	50	0.1	145	3.2	5.2	2.7	104	94
	132	108	7.0	3.1	3.9	19	0.6	134	23	56	<1	49	0.3	146	3.6	5.5	4.0	106	60
	133	137	7.4	3.4	4.0	20	0.6	96	28	57	1	79	0.2	146	3.7	5.7	4.2	104	61
	134	115	7.8	3.6	4.2	18	0.5	74	30	63	1	51	0.3	145	3.9	5.7	3.7	103	95
	135	106	7.2	3.2	4.0	17	0.6	124	28	62	<1	258	0.3	145	3.2	5.7	3.5	101	91
	136	98	6.1	3.0	3.1	18	0.7	128	25	100	<1	916	0.2	144	3.6	5.1	3.7	102	74
	137	116	7.7	3.4	4.3	22	0.7	77	26	52	<1	36	0.2	143	4.1	5.5	3.4	102	55
	138	124	7.1	3.3	3.8	19	0.7	70	28	70	<1	81	0.2	145	3.0	5.2	3.0	102	68
	139	109	7.7	3.5	4.2	16	0.7	96	68	143	<1	59	0.2	145	3.5	5.7	3.4	102	84
	140	129	7.2	3.3	3.9	19	0.6	117	53	72	<1	71	0.1	147	3.3	5.4	3.1	106	51
	Mean	114	7.2	3.3	3.9	19	0.6	102	39	79	<1	165	0.2	145	3.5	5.5	3.5	103	73
	sd	13.2	0.49	0.18	0.34	2.1	0.07	23.4	21.4	30.7		271.5	0.07	1.1	0.34	0.24	0.46	1.8	16.7
5F High dose	141	115	7.0	3.3	3.7	17	0.7	110	29	81	<1	113	0.2	147	3.7	5.5	4.6	106	58
	142	103	7.3	3.3	4.0	19	0.6	121	30	63	<1	50	0.2	148	3.4	5.7	4.6	105	107
	143	116	7.5	3.4	4.1	16	0.7	77	34	75	<1	122	0.2	147	3.5	5.7	4.5	105	84
	144	158	8.0	3.8	4.2	18	0.5	64	26	56	<1	112	0.2	147	3.7	5.7	3.6	104	104
	145	123	7.4	3.2	4.2	19	0.5	98	56	109	1	98	0.2	146	3.8	5.7	3.7	106	84
	146	133	7.4	3.4	4.0	18	0.7	102	26	70	1	122	0.1	145	3.7	5.3	3.8	103	64
	147	134	7.6	3.4	4.2	16	0.6	65	105	154	<1	79	0.2	146	3.7	5.7	3.1	105	84
	148	150	7.5	3.3	4.2	19	0.6	79	32	64	1	47	0.3	146	3.4	5.7	3.4	103	88
	149	96	7.0	3.1	3.9	17	0.7	121	22	59	<1	93	0.2	149	4.0	5.5	3.8	107	91
	150	123	7.6	3.5	4.1	22	0.7	83	26	54	<1	46	0.1	147	3.4	5.6	3.9	105	108
	Mean	125	7.4	3.4	4.1	18	0.6	92	39	79	<1	88	0.2	147	3.6	5.6	3.9	105	87
	sd	19.4	0.29	0.19	0.16	1.8	0.08	21.4	25.2	31.0		30.9	0.06	1.1	0.20	0.14	0.51	1.3	16.8

sd Standard deviation

APPENDIX 12

Urinalysis - individual values

Week 13 (5 November 1996)

Group	Animal no.	Volume ml	pH	SG	Protein mg/dl	TRS	Glucose	Ketones	Bile pigments	Urobilinogen	Haem pigments	Microscopy E	P	M	R	O	C	A
1M Air Control	1	9.4	7.3	1032	124	0	0	0	0	0	0	0	0	0	0	1	0	0
	2	4.4	6.9	1035	117	0	0	0	0	0	0	0	0	0	0	1	0	0
	3	6.4	6.9	1034	127	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	4	5.6	6.8	1028	128	0	0	tr	0	0	0	0	0	0	0	2	0	1sp
	5	2.6	6.2	1060	396	0	0	+	0	0	0	0	0	0	0	2	0	1sp
	6	3.6	6.9	1046	150	0	0	+	0	0	0	0	0	0	0	1	0	2sp
	7	2.4	6.8	1068	364	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	8	4.2	6.8	1044	150	0	0	tr	0	0	0	0	0	0	0	1	0	0
	9	2.0	6.5	1078	508	0	0	tr	0	0	0	0	0	0	0	1	0	0
	10	6.8	7.3	1031	130	0	0	tr	0	0	0	0	0	0	0	1	0	0
	Mean	4.7	6.8	1046	219													
	sd	2.33	0.33	17.4	145.1													
2M NaF	21	4.2	7.0	1042	143	0	0	tr	0	0	0	0	0	0	0	1	0	0
	22	3.0	7.2	1044	147	0	0	tr	0	0	0	0	0	0	0	1	0	0
	23	2.0	7.6	1066	132	0	0	++	0	0	0	0	0	0	0	1	0	1sp
	24	7.6	6.9	1037	127	0	0	+	0	0	+	0	0	0	0	1	0	1sp
	25	7.8	6.9	1032	447	0	0	0	0	0	0	0	1	0	0	1	0	0
	26	8.2	7.1	1028	121	0	0	+	0	0	0	0	0	0	1	1	0	1sp
	27	5.0	7.0	1039	128	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	28	2.6	6.8	1066	362	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	29	5.2	6.8	1043	146	0	0	+	0	0	0	0	0	0	0	1	0	2sp
	30	2.8	7.4	1049	352	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	Mean	4.8	7.1	1045	211													
	sd	2.33	0.26	12.8	124.5													

sd Standard deviation

APPENDIX 12

(Urinalysis - continued)

Week 13 (5 November 1996)

Group	Animal no.	Volume ml	pH	SG	Protein mg/dl	TRS	Glucose	Ketones	Bile pigments	Urobilinogen	Haem pigments	Microscopy E	P	M	R	O	C	A
3M Low dose	41	3.8	7.7	1046	149	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	42	4.2	7.1	1045	258	0	0	+	0	0	0	0	0	0	0	1	0	0
	43	5.2	7.4	1037	135	0	0	+	0	0	0	0	0	0	0	1	0	0
	44	5.8	7.3	1037	148	0	0	+	0	0	0	0	0	0	0	1	0	2sp
	45	8.0	7.6	1032	135	0	0	+	0	0	0	0	1	0	0	1	0	1sp
	46	3.0	7.9	1047	301	0	0	+	0	0	0	0	0	0	0	2	0	1sp
	47	3.6	7.1	1042	486	0	0	tr	0	0	0	0	0	0	0	1	0	0
	48	3.6	6.9	1044	147	0	0	tr	0	0	0	0	0	0	0	1	0	0
	49	7.8	7.3	1034	147	0	0	tr	0	0	0	0	0	0	0	1	0	0
	50	5.8	6.9	1040	140	0	0	tr	0	0	0	0	0	0	0	1	0	0
	Mean	5.1	7.3	1040	205													
	sd	1.77	0.34	5.2	114.6													
4M Inter dose	51	6.8	6.9	1030	127	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	52	5.4	7.3	1037	132	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	53	6.0	7.4	1031	122	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	54	6.0	7.4	1036	119	0	0	+	0	0	0	0	0	0	0	1	0	0
	55	10.5	7.6	1024	103	0	0	0	0	0	0	0	0	0	0	1	0	1sp
	56	3.4	7.5	1049	331	0	0	tr	0	0	0	0	0	0	0	1	0	2sp
	57	8.2	7.1	1028	111	0	0	tr	0	0	0	0	0	0	0	1	0	0
	58	10.2	7.3	1028	120	0	0	tr	0	0	0	0	0	0	0	1	0	2sp
	59	3.6	6.4	1056	358	0	0	0	0	0	0	0	1	0	0	1	0	1sp
	60	8.2	7.2	1032	102	0	0	tr	0	0	0	0	0	0	0	1	0	2sp
	Mean	6.8	7.2	1035	163													
	sd	2.45	0.35	10.1	96.6													

sd Standard deviation

APPENDIX 12

(Urinalysis - continued)

Week 13 (5 November 1996)

Group	Animal no.	Volume ml	pH	SG	Protein mg/dl	TRS	Glucose	Ketones	Bile pigments	Urobilinogen	Haem pigments	Microscopy E	P	M	R	O	C	A
5M High dose	61	6.2	7.5	1034	129	0	0	tr	0	0	0	0	0	0	0	2	0	1sp
	62	5.6	7.5	1034	120	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	63	4.0	7.1	1041	138	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	64	3.2	6.8	1046	146	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	65	4.6	6.8	1043	541	0	0	tr	0	0	0	0	0	0	0	1	0	0
	66	2.6	6.8	1066	417	0	0	tr	0	0	0	0	0	0	0	1	0	1sp
	67	6.0	7.6	1037	447	0	0	+	0	0	0	0	0	0	0	2	0	1sp
	68	2.4	8.0	1040	144	0	0	+	0	0	0	0	0	0	0	1	0	0
	69	4.2	6.6	1049	142	0	0	tr	0	0	0	0	0	0	0	2	0	1sp
	70	4.0	7.0	1027	276	0	0	+	0	0	0	0	0	0	0	1	0	1sp
	Mean	4.3	7.2	1042	250													
	sd	1.34	0.45	10.7	159.8													

sd Standard deviation

APPENDIX 12

(Urinalysis - continued)

Week 13 (5 November 1996)

Group	Animal no.	Volume ml	pH	SG	Protein mg/dl	TRS	Glucose	Ketones	Bile pigments	Urobilinogen	Haem pigments	Microscopy E	P	M	R	O	C	A
1F Air Control	81	3.2	7.6	1039	82	0	0	0	0	0	0	0	0	0	0	1	0	0
	82	2.0	6.4	1060	108	0	0	0	0	0	0	0	0	0	0	1	0	0
	83	2.2	6.2	1060	102	0	0	0	0	0	0	0	0	0	0	1	0	0
	84	3.6	6.0	1037	73	0	0	0	0	0	0	0	0	0	0	1	0	0
	85	2.6	6.0	1049	102	0	0	0	0	0	0	0	0	0	0	1	0	0
	86	5.8	6.4	1031	61	0	0	0	0	0	0	0	1	0	0	1	0	0
	87	3.2	6.4	1046	82	0	0	0	0	0	0	0	1	0	0	1	0	0
	88	3.0	6.6	1047	86	0	0	0	0	0	0	1	1	0	0	1	0	0
	89	2.6	6.4	1047	87	0	0	0	0	0	0	0	0	0	0	1	0	0
	90	3.6	6.3	1046	94	0	0	0	0	0	0	0	0	0	0	1	0	0
	Mean	3.2	6.4	1046	88													
	sd	1.07	0.45	9.2	14.4													
2F NaF	101	4.2	6.9	1036	72	0	0	0	0	0	0	0	0	0	0	1	0	0
	102	4.6	6.5	1036	60	0	0	0	0	0	0	0	1	0	0	1	0	0
	103	3.0	6.3	1050	83	0	0	0	0	0	0	0	0	0	0	1	0	0
	104	1.8	6.2	1068	105	0	0	0	0	0	0	0	0	0	0	1	0	0
	105	6.2	6.4	1029	54	0	0	0	0	0	0	0	1	0	0	1	0	0
	107	2.2	6.4	1048	89	0	0	0	0	0	0	0	0	0	0	1	0	0
	108	2.6	6.1	1049	91	0	0	0	0	0	0	0	0	0	0	1	0	0
	109	3.6	6.4	1037	72	0	0	0	0	0	0	0	0	0	0	1	0	0
	110	4.0	6.8	1036	72	0	0	0	0	0	0	0	0	0	0	1	0	0
	Mean	3.6	6.4	1043	78													
	sd	1.36	0.26	11.8	16.0													
3F Low dose	121	4.2	6.6	1034	65	0	0	0	0	0	0	0	0	0	0	2	0	0
	122	2.2	6.5	1064	81	0	0	0	0	0	0	0	0	0	0	1	0	0
	123	5.8	6.7	1033	59	0	0	0	0	0	0	0	1	0	0	1	0	0
	124	2.8	6.3	1048	84	0	0	0	0	0	0	0	1	0	0	1	0	0
	125	3.0	6.1	1034	102	0	0	0	0	0	0	0	0	0	0	1	0	0
	126	2.6	6.0	1039	80	0	0	0	0	0	0	0	0	0	0	1	0	0
	127	1.2	6.1	1062	99	0	0	0	0	0	0	0	0	0	0	1	0	0
	128	4.2	6.5	1038	65	0	0	0	0	0	0	1	0	0	0	1	0	0
	129	3.0	6.5	1044	77	0	0	0	0	0	0	0	1	0	0	1	0	0
	130	3.4	6.3	1042	71	0	0	0	0	0	0	0	0	0	0	1	0	0
	Mean	3.2	6.4	1044	78													
	sd	1.26	0.24	11.2	14.2													

sd Standard deviation

APPENDIX 12

(Urinalysis - continued)

Week 13 (5 November 1996)

Group	Animal no.	Volume ml	pH	SG	Protein mg/dl	TRS	Glucose	Ketones	Bile pigments	Urobilinogen	Haem pigments	Microscopy E	P	M	R	O	C	A
4F Inter dose	131	2.2	6.3	1060	89	0	0	0	0	0	0	0	0	0	0	1	0	0
	132	4.6	6.7	1038	65	0	0	0	0	0	0	0	1	0	0	1	0	0
	133	4.2	6.8	1037	66	0	0	0	0	0	0	0	0	0	0	1	0	0
	134	3.0	6.3	1041	113	0	0	0	0	0	0	0	0	0	0	1	0	0
	135	8.0	6.5	1027	69	0	0	0	0	0	0	0	1	0	0	1	0	0
	136	0.8	6.1	1062	105	0	0	0	0	0	0	0	0	0	0	1	0	0
	137	5.4	6.5	1032	63	0	0	0	0	0	0	0	0	0	0	1	0	0
	138	2.4	6.4	1042	110	0	0	0	0	0	0	0	0	0	0	1	0	0
	139	4.8	6.5	1035	59	0	0	0	0	0	0	0	0	0	0	1	0	0
	140	4.6	6.4	1046	76	0	0	0	0	0	0	0	0	0	0	1	0	0
	Mean	4.0	6.5	1042	82													
	sd	2.01	0.20	11.3	21.0													
5F High dose	141	2.2	6.5	1045	98	0	0	0	0	0	0	0	1	0	0	1	0	0
	142	3.8	6.6	1042	72	0	0	0	0	0	0	0	0	0	0	1	0	0
	143	2.0	6.4	1062	109	0	0	0	0	0	0	0	0	0	0	1	0	0
	144	2.8	6.2	1062	103	0	0	0	0	0	0	1	0	0	0	1	0	0
	145	2.4	6.4	1048	94	0	0	0	0	0	0	0	0	0	0	1	0	0
	146	6.2	6.6	1029	54	0	0	0	0	0	0	0	0	0	0	1	0	0
	147	6.0	6.6	1033	58	0	0	0	0	0	0	0	1	0	0	2	0	0
	148	3.8	6.3	1041	85	0	0	0	0	0	0	0	0	0	0	1	0	0
	149	5.4	6.4	1031	56	0	0	0	0	0	0	0	0	0	0	1	0	0
	150	3.2	6.1	1043	79	0	0	0	0	0	0	0	0	0	0	1	0	0
	Mean	3.8	6.4	1044	81													
	sd	1.57	0.17	11.5	20.3													

sd Standard deviation

APPENDIX 13

Urinary, bone and tooth fluoride and aluminium - pooled values at termination

Male rats - urine

		Concentration (mg/litre)	
Group	Cage	Fluoride	Aluminium
1 (Air control)	1	1.4	<0.5
	2	1.6	2.8
	Mean	1.5	1.5*
2 (NaF)	5	6.0	<0.5(<0.5)
	6	6.5	0.8
	Mean	6.25	0.5*
3 (Low dose Cryolite)	9	1.4(1.2)	<0.5
	10	1.1	<0.5
	Mean	1.25	<0.5
4 (Inter Dose Cryolite)	11	1.1(1.1)	<0.5
	12	1.7	0.9
	Mean	1.4	0.6*
5 (High dose Cryolite)	13	3.4(3.2)	<0.5
	14	4.2	3.4(3.0)
	Mean	3.8	1.8*

( ) Repeat analysis

* Calculation of mean, where levels <n included: $\frac{\Sigma n}{2}$+n/n(cage)

APPENDIX 13

(Urinary, bone and tooth fluoride and aluminium - pooled values at termination - continued)

Female rats - urine

Group	Cage	Concentration (mg/litre) Fluoride	Aluminium
1 (Air control)	17	1.5	<0.5
	18	1.2	<0.7
	Mean	1.35	<0.6
2 (NaF)	21	6.0	<0.5
	22	8.5	<0.6
	Mean	7.25	<0.6
3 (Low dose Cryolite)	25	1.4	1.8
	26	1.4	3.2
	Mean	1.4	2.5
4 (Inter dose Cryolite)	27	1.4	0.7
	28	1.6	1.5
	Mean	1.5	1.1
5 (High dose Cryolite)	29	5.5	<0.7
	30	3.7	4.9(4.6)
	Mean	4.6	2.6*

( ) Repeat analysis

* Calculation of mean, where levels <n included: $\frac{<n}{2}$+n/n(cage)

APPENDIX 13

(Urinary, bone and tooth fluoride and aluminium - pooled values at termination - continued)

Male rats - bone (femur)

Group	Cage	Concentration (% w/w) Fluoride	Aluminium
1 (Air control)	1	0.029	<0.01
	2	0.023	<0.01
	Mean	0.026	<0.01
2 (NaF)	5	0.053	<0.01
	6	0.060	<0.01
	Mean	0.057	<0.01
3 (Low dose Cryolite)	9	0.023	<0.01
	10	0.021	<0.01
	Mean	0.022	<0.01
4 (Inter dose Cryolite)	11	0.020	<0.01
	12	0.015 (0.018)	<0.01
	Mean	0.018	<0.01
5 (High dose Cryolite)	13	0.040	<0.01
	14	0.042	<0.01
	Mean	0.041	<0.01

( ) Repeat analysis

APPENDIX 13

(Urinary, bone and tooth fluoride and aluminium - pooled values at termination - continued)

Female rats - bone (femur)

		Concentration (% w/w)	
Group	Cage	Fluoride	Aluminium
1 (Air control)	17	0.031	<0.01
	18	0.033	<0.01
	Mean	0.032	<0.01
2 (NaF)	21	0.082 (0.086)	<0.01
	22	0.076 (0.090)	<0.01
	Mean	0.079	<0.01
3 (Low dose Cryolite)	25	0.027	<0.01(<0.01)
	26	0.032	<0.01
	Mean	0.030	<0.01
4 (Inter dose Cryolite)	27	0.025	<0.01
	28	0.031	<0.01
	Mean	0.028	<0.01
5 (High dose Cryolite)	29	0.063	<0.01
	30	0.063	<0.01
	Mean	0.063	<0.01

( ) Repeat analysis

APPENDIX 13

(Urinary, bone and tooth fluoride and aluminium - pooled values at termination - continued)

Male rats - teeth (lower incisors)

		Concentration (% w/w)	
Group	Cage	Fluoride	Aluminium
1 (Air control)	1	0.019	<0.01
	2	0.011	<0.01
	Mean	0.015	<0.01
2 (NaF)	5	0.019	<0.01
	6	0.020 (0.022)	<0.01
	Mean	0.020	<0.01
3 (Low dose Cryolite)	9	0.010	<0.01
	10	0.008	<0.01
	Mean	0.009	<0.01
4 (Inter Dose Cryolite)	11	0.017 (0.018)	<0.01
	12	0.016	<0.01
	Mean	0.017	<0.01
5 (High dose Cryolite)	13	0.017	<0.01
	14	0.017	<0.01
	Mean	0.017	<0.01

( ) Repeat analysis

APPENDIX 13

(Urinary, bone and tooth fluoride and aluminium - pooled values at termination - continued)

Female rats -teeth (lower incisors)

Group	Cage	Concentration (% w/w)	
		Fluoride	Aluminium
1 (Air control)	17	0.016	<0.01 (<0.01)
	18	0.020	<0.01
	Mean	0.018	<0.01
2 (NaF)	21	0.025	<0.01
	22	0.010	<0.01
	Mean	0.018	<0.01
3 (Low dose Cryolite)	25	0.012	<0.01
	26	0.013	<0.01
	Mean	0.013	<0.01
4 (Inter Dose Cryolite)	27	0.018	<0.01
	28	0.018	<0.01
	Mean	0.018	<0.01
5 (High dose Cryolite)	29	0.022	<0.01
	30	0.024	<0.01 (<0.01)
	Mean	0.023	<0.01

( ) Repeat analysis

APPENDIX 14

Urinary, bone and tooth fluoride and aluminium - pooled values at withdrawal

Male rats - urine

Group	Cage	Concentration (mg/litre)	
		Fluoride	Aluminium
1 (Air control)	3	1.0	<0.5 (<0.5) (0.6)
	4	1.1	<0.05
	Mean	1.05	<0.5
2 (NaF)	7	1.6	<0.5
	8	1.4	<0.5 (<0.5)
	Mean	1.5	<0.5
5 (High dose Cryolite)	15	0.9	<0.5
	16	1.0 (1.0)	<0.5
	Mean	0.95	<0.5

( ) Repeat analysis

APPENDIX 14

(Urinary, bone and tooth fluoride and aluminium - pooled values at withdrawal - continued)

Female rats - urine

Group	Cage	Concentration (mg/litre)	
		Fluoride	Aluminium
1 (Air control)	19	1.2	<0.5
	20	0.96 (0.91)	<0.5
	Mean	1.08	<0.5
2 (NaF)	23	1.3	0.8
	24	1.3	<0.6
	Mean	1.3	0.24
5 (High dose Cryolite)	31	1.2	<0.5
	32	1.2	<0.5
	Mean	1.2	<0.5

( ) Repeat analysis

APPENDIX 14

(Urinary, bone and tooth fluoride and aluminium - pooled values at withdrawal - continued)

Male rats - bone (femur)

Group	Cage	Concentration (% w/w)	
		Fluoride	Aluminium
1 (Air control)	3	0.027	<0.01
	4	0.026	<0.01 (<0.1)
	Mean	0.027	<0.01
2 (NaF)	7	0.058	<0.01
	8	0.050	<0.01
	Mean	0.054	<0.01
5 (High dose Cryolite)	15	0.038 (0.038)	<0.01
	16	0.040	<0.01 (<0.1)
	Mean	0.039	<0.01

( ) Repeat analysis

APPENDIX 14

(Urinary, bone and tooth fluoride and aluminium - pooled values at withdrawal - continued)

Female rats - bone (femur)

Group	Cage	Concentration (% w/w)	
		Fluoride	Aluminium
1 (Air control)	19	0.036	<0.01
	20	0.038	<0.01
	Mean	0.037	<0.01
2 (NaF)	23	0.075	<0.01
	24	0.070	<0.01
	Mean	0.073	<0.01
5 (High dose Cryolite)	31	0.064 (0.064)	<0.01
	32	0.051	<0.01
	Mean	0.064	<0.01

( ) Repeat analysis

Rat 151 Group 5 - sacrificed 1 November 1996 (Week 12)	0.052	<0.01

APPENDIX 14

(Urinary, bone and tooth fluoride and aluminium - pooled values at withdrawal - continued)

Male rats - teeth (lower incisors)

		Concentration (% w/w)	
Group	Cage	Fluoride	Aluminium
1 (Air control)	3	0.014	<0.01
	4	0.012	<0.01
	Mean	0.013	<0.01
2 (NaF)	7	0.011	<0.01
	8	0.010	<0.01(<0.01)
	Mean	0.011	<0.01
5 (High dose Cryolite)	15	0.009	<0.01
	16	0.010	<0.01
	Mean	0.010	<0.01

( ) Repeat analysis

APPENDIX 14

(Urinary, bone and tooth fluoride and aluminium - pooled values at withdrawal - continued)

Female rats - teeth (lower incisors)

		Concentration (% w/w)	
Group	Cage	Fluoride	Aluminium
1 (Air control)	19	0.015	<0.01
	20	0.016 (0.017)	<0.01
	Mean	0.016	<0.01
2 (NaF)	23	0.012	<0.01
	24	0.014	<0.01
	Mean	0.013	<0.01
5 (High dose Cryolite)	31	0.014	<0.01
	32	0.011	<0.01 (<0.01)
	Mean	0.013	<0.01
Rat 151 Group 5 - sacrificed 1 November 1996 (Week 12)		0.028	<0.01

( ) Repeat analysis

APPENDIX 15

Organ weights - individual values - termination

Week 14

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg	Testes L g	Testes R g	Epididymides Left g	Epididymides Right g
1M Air Control	1	468	1.44	13.9	2.25	49.2	1.72	1.79	0.576	0.609
	2	373	1.24	11.7	1.98	54.7	1.65	1.43	0.544	0.425
	3	439	1.37	14.5	2.40	63.5	1.98	1.82	0.614	0.559
	4	436	1.50	13.7	2.61	54.0	1.57	1.66	0.564	0.606
	5	414	1.77	13.6	2.59	43.0	1.89	1.80	0.644	0.613
	6	407	1.50	13.3	2.28	49.7	1.74	1.75	0.569	0.560
	7	379	1.37	12.8	2.31	40.7	1.67	1.66	0.592	0.612
	8	462	1.46	14.6	2.66	52.1	1.70	1.65	0.607	0.623
	9	482	1.63	13.9	2.70	49.1	1.75	1.78	0.640	0.595
	10	392	1.33	11.2	2.06	61.9	1.57	1.51	0.577	0.612
	Mean	425	1.46	13.3	2.38	51.8	1.72	1.69	0.593	0.581
	sd	38.1	0.153	1.12	0.252	7.24	0.128	0.131	0.0330	0.0592
2M NaF	21	455	1.56	15.5	2.80	65.7	1.93	1.93	0.579	0.605
	22	351	1.18	10.5	2.01	56.2	1.66	1.55	0.595	0.525
	23	394	1.39	11.5	2.18	47.7	1.68	1.54	0.512	0.537
	24	454	1.75	13.4	2.59	77.1	1.65	1.68	0.606	0.620
	25	548	1.60	18.2	3.26	81.6	1.83	1.83	0.730	0.725
	26	454	1.67	14.7	2.59	49.9	2.00	2.10	0.664	0.683
	27	407	1.30	11.2	2.61	52.8	1.65	1.40	0.580	0.526
	28	429	1.38	11.9	2.36	60.3	1.76	1.85	0.644	0.682
	29	479	1.67	13.5	2.98	59.9	1.66	1.71	0.617	0.663
	30	436	1.28	13.4	2.13	63.6	1.84	1.75	0.640	0.633
	Mean	441	1.48	13.4	2.55	61.5	1.77	1.73	0.617	0.620
	sd	52.8	0.195	2.32	0.394	11.07	0.129	0.206	0.0584	0.0714

sd Standard deviation

APPENDIX 15

(Organ weights - continued)

Week 14

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg	Testes L g	Testes R g	Epididymides Left g	Epididymides Right g
3M Low dose	41	409	1.18	11.1	2.50	49.4	1.78	1.66	0.604	0.581
	42	413	1.44	11.7	2.61	39.0	1.45	1.35	0.532	0.531
	43	456	1.44	15.6	3.15	94.3	1.70	1.69	0.560	0.596
	44	456	1.59	15.6	2.79	52.0	1.71	1.68	0.638	0.624
	45	462	1.70	13.9	2.66	53.1	1.75	1.66	0.666	0.615
	46	402	1.29	13.0	2.34	41.9	1.73	1.66	0.497	0.484
	47	365	1.31	12.0	2.29	53.2	1.64	1.32	0.506	0.511
	48	428	1.49	13.2	2.21	44.4	1.70	1.60	0.602	0.547
	49	567	1.74	20.3	3.42	56.7	1.81	1.72	0.587	0.570
	50	412	1.49	13.9	2.45	54.4	1.66	1.66	0.622	0.578
	Mean	437	1.47	14.0	2.64	53.8	1.69	1.60	0.581	0.564
	sd	54.4	0.178	2.67	0.389	15.35	0.099	0.144	0.0565	0.0451
4M Inter dose	51	483	1.46	13.9	2.71	51.4	1.83	1.76	0.675	0.629
	52	436	1.45	15.3	2.55	54.4	1.35	1.46	0.591	0.571
	53	460	1.50	14.5	2.86	75.8	1.68	1.72	0.601	0.590
	54	452	1.63	14.6	2.63	58.3	1.68	1.59	0.635	0.529
	55	434	1.53	14.6	2.40	52.6	1.61	1.61	0.521	0.505
	56	444	1.75	12.6	2.63	38.7	1.76	1.65	0.628	0.587
	57	404	1.53	12.5	2.52	38.2	1.59	1.36	0.515	0.436
	58	431	1.45	13.2	2.33	55.6	1.96	1.89	0.640	0.616
	59	424	1.41	12.5	2.54	53.4	1.58	1.42	0.549	0.455
	60	470	1.55	14.4	2.66	60.2	1.37	1.31	0.524	0.565
	Mean	444	1.52	13.8	2.58	53.9	1.64	1.58	0.588	0.548
	sd	23.2	0.100	1.03	0.153	10.68	0.187	0.187	0.0574	0.0656

sd Standard deviation

APPENDIX 15

(Organ weights - continued)

Week 14

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg	Testes L g	Testes R g	Epididymides Left g	Epididymides Right g
5M High dose	61	450	1.73	15.2	2.44	51.6	1.72	1.57	0.577	0.575
	62	357	1.69	11.8	2.34	62.6	1.62	1.61	0.438	0.509
	63	378	1.45	12.0	2.59	45.9	1.59	1.65	0.472	0.502
	64	414	1.52	13.3	2.50	44.7	1.65	1.58	0.606	0.564
	65	508	2.04	14.0	2.75	73.1	1.74	1.80	0.629	0.688
	66	457	1.57	15.0	2.28	48.6	1.63	1.56	0.461	0.913
	67	426	1.59	13.9	2.68	59.1	1.57	1.54	0.489	0.464
	68	393	1.51	13.7	2.33	48.5	1.79	1.75	0.584	0.561
	69	451	1.57	15.6	2.97	51.2	1.82	1.62	0.591	0.509
	70	413	2.08	14.3	2.46	56.7	1.76	1.73	0.620	0.518
	Mean	425	1.68	13.9	2.53	54.2	1.69	1.64	0.547	0.580
	sd	44.0	0.218	1.27	0.216	8.80	0.090	0.089	0.0730	0.1319

sd Standard deviation

APPENDIX 15

(Organ weights - continued)

Week 14

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg
1F Air Control	81	228	1.11	7.6	1.53	49.6
	82	244	1.09	8.8	1.57	66.7
	83	256	1.11	8.4	1.66	67.5
	85	276	1.13	9.7	1.80	62.6
	86	256	1.01	8.5	1.61	57.7
	87	248	1.14	8.2	1.67	50.6
	88	275	1.10	9.5	1.86	64.6
	89	274	1.13	9.9	1.65	73.9
	90	288	1.21	9.7	1.92	70.6
	Mean	261	1.12	8.9	1.70	62.6
	sd	19.2	0.054	0.82	0.134	8.47
2F NaF	101	250	1.09	8.2	1.60	75.6
	102	282	1.19	9.1	1.76	40.7
	103	225	1.04	8.7	1.53	62.0
	104	247	1.03	8.0	1.64	53.9
	105	255	1.27	7.7	1.70	64.0
	107	245	1.20	8.6	1.71	70.2
	108	282	1.31	9.7	1.82	77.7
	109	264	1.13	8.0	1.80	68.7
	110	263	1.09	9.7	1.68	71.8
	Mean	257	1.15	8.6	1.69	65.0
	sd	18.1	0.098	0.74	0.092	11.64
3F Low dose	121	233	1.21	8.1	1.65	93.1
	122	259	1.05	8.5	1.59	53.2
	123	270	1.15	8.3	1.76	73.5
	124	279	1.22	10.0	1.96	84.5
	125	228	1.38	10.5	1.81	66.7
	126	276	1.11	10.2	1.93	70.7
	127	233	1.10	7.6	1.43	64.0
	128	244	1.16	8.2	1.47	61.0
	129	228	1.19	8.4	1.62	79.3
	130	249	1.09	8.1	1.61	58.8
	Mean	250	1.17	8.8	1.68	70.5
	sd	20.1	0.094	1.03	0.179	12.37

sd Standard deviation

APPENDIX 15

(Organ weights - continued)

Week 14

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg
4F Inter dose	131	256	1.27	8.4	1.90	66.7
	132	227	1.04	6.8	1.46	50.7
	133	219	0.95	7.0	1.63	68.4
	134	219	0.92	8.3	1.80	67.0
	135	323	1.33	10.4	1.95	86.0
	136	233	1.26	8.1	1.47	75.8
	137	246	1.32	9.8	1.59	68.1
	138	274	1.36	10.0	2.14	70.9
	139	250	1.15	8.3	1.48	64.3
	140	276	1.25	9.7	1.85	93.7
	Mean	252	1.18	8.7	1.73	71.2
	sd	32.1	0.162	1.26	0.235	11.88
5F High dose	141	226	1.27	7.4	1.64	67.5
	142	254	1.30	8.3	1.52	55.0
	143	294	1.45	9.5	1.91	80.2
	144	245	1.25	9.1	1.65	55.7
	145	220	1.21	8.0	1.60	66.2
	146	266	1.29	8.9	1.49	65.6
	147	287	1.31	10.2	1.79	59.9
	148	279	1.26	9.0	1.48	73.5
	149	240	1.30	8.0	1.34	69.5
	150	230	1.11	8.7	1.32	69.0
	Mean	254	1.27	8.7	1.57	66.2
	sd	26.2	0.085	0.84	0.186	7.78

sd Standard deviation

APPENDIX 16

Organ weights - individual values - withdrawal

Week 27

Group	Animal no.	Body wt	Lungs	Liver	Kidneys	Adrenals	Testes L	Testes R	Epididymides Left	Epididymides Right
		g	g	g	g	mg	g		g	g
1M Air Control	11	608	1.85	17.5	3.31	55.7	1.85	1.85	0.736	0.822
	12	529	1.92	16.4	3.26	53.0	1.92	1.96	0.704	0.701
	13	590	2.12	18.2	3.19	51.6	2.02	1.98	0.701	0.774
	14	534	1.80	15.7	2.61	40.6	1.44	1.45	0.559	0.570
	15	578	1.75	15.9	2.69	49.5	1.89	1.84	0.636	0.644
	16	604	1.86	16.9	3.11	55.1	1.93	1.90	0.673	0.740
	17	687	2.03	19.7	3.51	61.9	1.85	1.93	0.737	0.736
	18	591	1.68	17.6	3.01	49.8	1.74	1.75	0.702	0.821
	19	579	1.40	16.7	2.57	44.8	1.56	1.55	0.488	0.511
	20	642	1.58	19.7	3.47	48.8	1.60	1.65	0.581	0.735
	Mean	594	1.80	17.4	3.07	51.1	1.78	1.79	0.652	0.705
	sd	46.8	0.211	1.42	0.345	5.94	0.188	0.182	0.0838	0.1025
2M NaF	31	638	1.80	20.7	3.29	49.2	1.96	1.97	0.767	0.827
	32	450	1.74	13.3	2.60	34.3	1.69	1.79	0.661	0.681
	33	603	2.03	22.7	2.94	47.9	1.86	1.85	0.706	0.741
	34	634	2.40	21.5	3.43	55.0	1.90	1.83	0.691	0.736
	35	588	2.25	17.7	2.90	55.1	1.94	1.84	0.655	0.661
	36	601	2.11	18.0	3.32	57.2	2.04	2.02	0.806	0.780
	37	622	1.89	17.5	3.10	61.9	1.95	1.97	0.775	0.749
	38	742	2.61	23.6	4.01	91.4	1.85	1.79	0.677	0.686
	39	544	1.82	16.6	3.43	61.5	1.55	1.61	0.663	0.679
	40	654	1.73	18.9	3.41	49.1	1.56	1.56	0.586	0.618
	Mean	608	2.04	19.1	3.24	56.3	1.83	1.82	0.699	0.716
	sd	75.5	0.301	3.11	0.385	14.72	0.172	0.150	0.0666	0.0621

sd Standard deviation

APPENDIX 16

(Organ weights - continued)

Week 27

Group	Animal no.	Body wt	Lungs	Liver	Kidneys	Adrenals	Testes L	Testes R	Epididymides Left	Epididymides Right
		g	g	g	g	mg	g		g	g
5M High dose	71	504	1.60	16.7	2.79	45.5	1.80	1.79	0.706	0.690
	72	537	2.02	15.5	2.84	55.0	1.84	1.83	0.701	0.735
	73	598	1.86	18.4	3.53	47.8	1.66	1.84	0.645	0.703
	74	558	2.44	17.8	3.46	60.6	2.04	2.01	0.733	0.731
	75	640	2.44	21.2	4.04	56.1	1.74	1.72	0.673	0.625
	76	511	2.12	14.6	2.68	61.9	1.73	1.77	0.640	0.614
	77	551	2.24	16.9	3.20	50.3	1.89	1.90	0.772	0.695
	78	591	1.85	17.5	3.28	52.7	1.77	1.80	0.703	0.694
	79	550	2.25	14.6	3.06	47.1	1.98	1.90	0.677	0.709
	80	527	1.93	20.4	3.21	53.4	2.05	2.01	0.676	0.681
	Mean	557	2.07	17.4	3.21	53.0	1.85	1.86	0.693	0.688
	sd	42.0	0.272	2.22	0.406	5.54	0.137	0.097	0.0398	0.0399

sd Standard deviation

APPENDIX 16

Organ weights - individual values

Week 27

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg
1F Air Control	91	387	1.64	13.9	2.35	81.0
	92	322	1.53	11.0	2.09	66.7
	93	319	1.50	10.4	1.95	91.9
	94	338	1.32	13.2	2.15	92.1
	95	#0	#1.33	#11.9	#2.09	#76.1
	96	343	1.30	11.9	2.14	75.7
	97	301	1.22	10.5	1.83	50.9
	98	334	1.57	14.0	2.52	99.8
	99	295	1.41	11.5	1.93	66.1
	100	268	1.11	9.3	1.73	59.0
	Mean	323	1.40	11.7	2.08	75.9
	sd	33.8	0.176	1.65	0.250	16.61
2F NaF	111	312	1.41	12.0	2.06	65.9
	112	289	1.53	12.4	2.03	73.8
	113	371	1.54	11.2	2.04	62.7
	114	309	1.76	12.5	1.88	72.2
	115	288	1.71	11.3	2.26	79.7
	116	288	1.24	11.0	1.95	75.7
	117	281	1.25	11.0	1.86	56.7
	118	341	1.46	13.1	1.92	69.9
	119	317	1.44	11.4	1.80	57.9
	120	304	1.32	10.4	1.85	58.0
	Mean	310	1.47	11.6	1.96	67.3
	sd	27.9	0.177	0.83	0.135	8.22

sd Standard deviation

\# Value excluded from calculation of mean and statistical analysis
Bodyweight not recorded, in error, at necropsy

APPENDIX 16

(Organ weights - continued)

Week 27

Group	Animal no.	Body wt g	Lungs g	Liver g	Kidneys g	Adrenals mg
5F High dose	152	314	1.38	11.0	2.10	58.2
	153	329	1.64	12.8	2.26	57.9
	154	347	1.68	13.1	2.38	66.4
	155	335	1.35	10.4	2.06	68.9
	156	305	1.86	10.9	2.17	88.2
	157	311	1.63	11.0	2.07	68.5
	158	308	1.53	9.0	1.84	66.9
	159	319	1.45	11.9	1.97	60.5
	160	334	1.48	11.7	1.90	88.3
	Mean	321	1.55	11.3	2.08	69.3
	sd	13.9	0.161	1.25	0.172	11.53

sd Standard deviation

APPENDIX 17

Preparation of rat bone marrow metaphase slides

SUMMARY

The objective of this part of the study was to prepare slides for the assessment of clastogenic potential of Sodium hexafluoroaluminate (Cryolite) in an *in vivo* cytogenetic test system using the satellite animals.

Two hours prior to sacrifice, all satellite group rats were treated with a single intraperitoneal dose of colchicine to arrest dividing cells at metaphase. Six satellite male rats in the negative control, the sodium fluoride treated group and the test substance treated group were sacrificed 24 hours after completion of treatment. Six male rats dosed with a single intraperitoneal injection of cyclophosphamide at 25 mg/kg in the positive control satellite group were sacrificed 24 hours after dosing.

Both femurs were dissected from each satellite animal and the bone marrow was aspirated with Hanks' balanced salts solution then treated with a hypotonic potassium chloride solution. The cells were fixed and a homogenous cell suspension was prepared. At least four slides were prepared from each animal. Two of these slides from each animal were stained in Giemsa.

Two stained and two unstained slides from each animal were despatched to Bayer AG for analysis of chromosomal aberrations. The results of this analysis are not presented in this report and remain the responsibility of the Sponsor.

INTRODUCTION

Sodium hexafluoroaluminate (Cryolite) was tested for potential clastogenicity in the rat bone marrow metaphase test. The protocol was designed to comply with the following guidelines;

OECD Short-term Toxicology Group (Guideline No. 475)
EEC Directive 79/831 EEC Annex V (Method B.11)

Chemical mutagens act by disrupting the genetic material of the cell (*ie* DNA). Most mutagens will cause lesions in DNA leading to production of chromosomal aberrations. Such chromosomal damage is best viewed at the metaphase stage of cell division when the chromosomes are most clearly visible. Chromosomal aberrations are evaluated at the first post-treatment metaphase after treatment with the test substance since induced damage may subsequently be repaired or may result in death of the cell.

Cytogenetic tests can be carried out using cultured cells but the main disadvantages of *in vitro* cytogenetic tests are that metabolic processes, distribution and excretion are not reflected in such tests.

APPENDIX 17

(Preparation of rat bone marrow metaphase slides - continued)

In this study, rats were treated with the test substance by inhalation which is the major or most significant expected route of human exposure. Rats are usually chosen for cytogenetic tests in accordance with regulatory guidelines, because a large volume of test data is available for this species, pure-bred strains are readily available, and the chromosomes of the rat are easily analyzed. The strain was selected in view of the availability of comprehensive background data, relating to clinical and pathological parameters, at this laboratory. The rapidly dividing cells of the young animal's bone marrow are especially sensitive to DNA damage and the high rate of cell division also facilitates analysis.

EXPERIMENTAL PROCEDURE

POSITIVE CONTROL COMPOUND

Identity:	Cyclophosphamide (Batch 26H0473)
Supplier:	Sigma Chemical Co. Ltd.
Appearance:	White powder
Purity:	>98%
Solubility:	Freely soluble
Storage:	*c.* 4°C desiccated in darkness
Vehicle:	0.92% saline (obtained from Baxters batch number 96I13B27)
Dose preparation:	Solution prepared just prior to use at a concentration of 2.5 mg/ml

EXPERIMENTAL DESIGN

Satellite Group/Colour code	Material	Dosage	Sampling time †	Animal number
1:White	Air	-	24	161 - 166
2:white/black	Sodium fluoride	6 mg/m^3	24	167 - 172
5:Red/black	Sodium hexafluoroaluminate	5 mg/m^3	24	173 - 178
6:Grey/black	Cyclophosphamide	30 mg/kg	24	179 - 184

† Hours after completion of exposure/dosing

APPENDIX 17

(Preparation of rat bone marrow metaphase slides - continued)

ANIMAL TREATMENT

The animals in the positive control group (Group 6) were treated with a single intraperitoneal injection of cyclophosphamide at a dosage of 25 mg/kg using a standard dose volume of 10 ml/kg bodyweight. The dosing was performed so as to coincide with the time of completion of the inhalation exposure.

Following completion of the inhalation exposure and dosing of the positive control group, and prior to colchicine administration, all satellite group animals were transferred to polystyrene disposable cages fitted with stainless steel mesh lids and taken to the Department of Cellular Toxicology, Building C31.

Animals were weighed prior to treatment and just prior to sacrifice. Bodyweights ranged from 395 to 555 grams at sacrifice. No clinical signs or mortalities were observed at any time after completion of treatment.

PREPARATION OF BONE MARROW METAPHASE SLIDES

Approximately 2½ hours prior to sacrifice, all satellite animals were treated with colchicine by intraperitoneal injection at a dose level of 4 mg/kg to arrest dividing cells at metaphase. Colchicine was obtained from Sigma Chemical Co. Ltd (approximately 95% pure) and was formulated on the day of use as a solution in 0.9% saline (obtained from Baxters, batch number 96120B26) at a concentration of 0.4 mg/ml.

Approximately 24 hours after completion of inhalation exposure and after dosing Group 6, six male rats from each group were killed by cervical dislocation. Both femurs were dissected from each animal. The distal heads were kept intact and as much tissue as possible was removed from the bones.

For each femur, a Sterilin universal bottle labelled with the animal number containing 10 ml Hanks' Balanced Salts solution was prepared. A 5 ml syringe fitted with a 21 g needle was inserted into the femur, and the bone marrow together with the Hanks' Balanced Salts solution from the relevant bottle was drawn up into the syringe and then ejected back into the bottle. The bone was discarded and the cells were further aspirated through the needle. The cells were centrifuged at 1000 rpm for 5 minutes. The supernatant was discarded, the cell pellet was knocked up and resuspended in 10 ml of freshly-prepared, hypotonic potassium chloride solution (0.56% KCl obtained from Fisons, batch number 108552A314, formulated in distilled water). The cells were incubated in this hypotonic solution for 10 minutes at room temperature to enhance the eventual spreading of the metaphase chromosomes by swelling the cells.

APPENDIX 17

(Preparation of rat bone marrow metaphase slides - continued)

The cells were then centrifuged for 5 minutes at 1000 rpm. The supernatant was discarded and the cells were resuspended in approximately 1 ml of hypotonic solution. The cells were then fixed by the gradual addition of approximately 9 ml of freshly-prepared, ice-cold fixative (3:1 (v/v) methanol:glacial acetic acid) whilst stirring the cells on a rotary mixer. The fixed cell suspensions were then stored at approximately 4°C for at least 24 hours prior to further processing.

The cells were resuspended in the fixative by vigorous stirring and then centrifuged at 1500 rpm for 10 minutes. The supernatant was discarded and the cells were resuspended in 10 ml freshly-prepared ice-cold fixative. The cells were then centrifuged at 1500 rpm for 10 minutes, the supernatant discarded and the cells were resuspended in approximately 0.5 ml of fixative. The cells were dispersed to produce a suspension using a pasteur pipette.

Two or three drops of the final cell suspensions were dropped onto pre-chilled microscope slides labelled with the study number and animal number. At least four slides were prepared from each animal. They were placed in a dust-free container and left to air-dry at room temperature.

FIXATION AND STAINING OF SLIDES

Two of the slides from each animal were rinsed in purified water and then stained for 20 minutes in 10% Giemsa stain (freshly prepared by dilution of 100 ml of Giemsa's improved R66 solution (BDH) with 900 ml purified water then filtered through cotton wool). The slides were differentiated in pH 6.8 buffered distilled water for 10 minutes, air-dried and mounted with coverslips using DPX mountant which was allowed to harden at approximately 37°C.

These slides together with two unstained slides from each animal were despatched, with prior notification, to the Sponsor's authorised delegate Dr B A Herbold of Bayer AG, Institute of Toxicology, Carcinogenicity and Genotoxicity, Friedrich-Ebert Str 217-333, D-42096 Wuppertal, Germany, for microscopic examination. Dr A Herbold is the responsible scientist. The results of this analysis are not provided in this report and remain the responsibility of BG Chemie. The archiving and storage of slides and is the responsibility of the Sponsor.

APPENDIX 17

(Preparation of rat bone marrow metaphase slides - continued)

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APPENDIX 18

Individual clinical and pathological findings

In this appendix the macroscopic and microscopic findings relating to each animal are listed.

The initial examination was undertaken by the study pathologist, the results of which were then subjected to a routine peer review by a second pathologist. The diagnoses reported here represent the consensus opinions of both pathologists.

Study Pathologist: Richard L Gregson, M.Phil., Ph.D., C.Biol., M.I.Biol.,
Senior Pathologist,
Department of Pathology.

Peer Review: David J Lewis, Ph.D., F.R.C.Path.,
Consultant Pathologist,
Department of Pathology.

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 1M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - arytenoids: (Minimal)

Lungs
Aggregates of macrophages: (Trace)

Thymus
Involution/atrophy: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 2M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Testes
Atrophy: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Air
Dosage Level:	Control
Rat No/Sex:	3M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Heart
Myocarditis: (Minimal)

Thymus
Involution/atrophy: (Trace)

Lymph Nodes - Mediastinal
Sinus congestion with siderocytes: (Moderate)

Testes
Atrophy: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 4M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Heart
Myocardial necrosis: (Minimal)

Liver
Portal inflammation: (Minimal , Focus)

Kidneys
Tubular basophilia: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 5M (Terminal)

MACROSCOPIC FINDINGS

Lungs
Congested: (Minimal)

Liver
Median cleft, pale subcapsular area/s: (One) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Recent alveolar haemorrhage: (Minimal , Focal)

Spleen
Capsular inflammation: (Minimal)

Liver
Vacuolated hepatocytes at median cleft

Eyes
Retinal rosettes: (Minimal , Unilateral)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 6M (Terminal)

MACROSCOPIC FINDINGS

Lungs
Congested: (Minimal)
Pale subpleural foci: (A few , Punctate) right posterior lobe

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Nasal Passages
Degeneration of olfactory epithelium: (Minimal , Focal)

Lungs
Subpleural aggregation of alveolar macrophages: (Minimal)

The following tissues were considered normal:

Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 7M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Heart
Myocarditis: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 8M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Heart
Myocarditis: (Trace)

Liver
Parenchymal inflammatory cells: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 9M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 10M (Terminal)

MACROSCOPIC FINDINGS

Lungs
Congested: (Patchy)

Liver
Median cleft, pale subcapsular area/s: (One) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Vascular congestion: (Trace)

Heart
Myocarditis: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver : (W.N.L.); Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 11M (Recovery)

MACROSCOPIC FINDINGS

Skin Scabs
Side/s of face: (Right , One) 1mm

Skin Alopecia
Cranial region
Side/s of face

Lymph Nodes - Cervical
Congested

Lungs
Pale subpleural foci: (Left , A few) 1mm

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonia: (Minimal , Foci/areas)

Lymph Nodes - Cervical
Increased cellularity - generalised: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Skin
Epidermal erosion: (Moderate)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 11M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 12M (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cranial region: (Diffuse)

Lymph Nodes - Mediastinal
Enlarged: 3mm

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Lymph Nodes - Mediastinal
Increased lymphoid cellularity - generalised: (Minimal)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 13M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested: (Minimal)
Pale subpleural foci: (Multiple , Punctate)

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

Liver
Median cleft, pale subcapsular area/s: (One) 2mm

Forestomach
A caseous cyst: 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Stomach
Cysts at limiting ridge: (Moderate)

The following tissues were considered normal:

Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 14M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 15M (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cranial region: (Diffuse)

Lungs
Congested

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)
Aggregates of macrophages: (Trace , Focal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 16M (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Side/s of face: (Minimal)

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Moderate)
Perivascular inflammatory infiltration: (Trace)
Granulomatous inflammation: (Moderate , Foci/areas)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Tissues not available for examination were:

Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 17M (Recovery)

MACROSCOPIC FINDINGS

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 18M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 19M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace , Area , One lobe)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 20M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial

Tissues not available for examination were:

Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 21M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Necrosis of ventral cartilage
Epithelial hyperplasia - ventral: (Minimal)

Lungs
Pneumonitis: (Trace)
Subpleural aggregation of alveolar macrophages: (Trace)

Lymph Nodes - Cervical
Sinus histiocytosis: (Minimal)

Kidneys
Pelvic suburothelial inflammatory cells: (Moderate , Unilateral)

Testes
Atrophy: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 21M - continued

MICROSCOPIC FINDINGS - continued

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 22M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Moderate)

Lungs
Eosinophilic crystals assoc. macrophages around alveolar ducts: (Minimal)

Heart
Myocarditis: (Trace)

Testes
Atrophy: (Minimal , Unilateral)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Tissues not available for examination were:

Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 23M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Trace)

Liver
Parenchymal inflammatory cells: (Minimal)

Kidneys
Tubular basophilia: (Trace)

Urinary Bladder
Serosal inflammation with siderocytes: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Pancreas; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 24M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Moderate)
Subepithelial inflammation - ventral: (Minimal)

Lungs
Aggregates of macrophages: (Trace)

Heart
Myocarditis: (Trace)

Kidneys
Dystrophic mineralisation: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Tissues not available for examination were:

Parathyroids : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 25M (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Side/s of face: (Minimal , Diffuse)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)
Subepithelial inflammation - ventral: (Trace)

Lungs
Aggregates of macrophages: (Trace)

Heart
Myocarditis: (Minimal)

Lymph Nodes - Mediastinal
Sinus histiocytosis with mast cells: (Moderate)

Kidneys
Tubular basophilia: (Minimal)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 25M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 26M (Terminal)

MACROSCOPIC FINDINGS

Tail
Malaligned

Lungs
Pale subpleural foci: (A few , Punctate)

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Trace)

Lungs
Aggregates of macrophages: (Minimal)

Parathyroids
Cyst

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 26M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 27M (Terminal)

MACROSCOPIC FINDINGS

Kidneys
Increased pelvic dilatation: (Left , Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)
Subepithelial inflammation - ventral: (Trace)

Spleen
Haemosiderosis: (Minimal)

Kidneys
Dilatation of the renal pelvis: (Minimal , Unilateral)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 28M (Terminal)

MACROSCOPIC FINDINGS

Lymph Nodes - Cervical
Congested: (Minimal)

Liver
Median cleft, pale subcapsular area/s: (One) 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Moderate)
Epithelial mineralisation - ventral: (Moderate)

Lungs
Aggregates of macrophages: (Minimal)

Heart
Myocarditis: (Trace)

Lymph Nodes - Cervical
Increased cellularity - generalised: (Minimal)

Kidneys
Tubular basophilia: (Minimal)
Dilatation of the renal pelvis: (Minimal , Unilateral)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 28M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver : (W.N.L.); Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 29M (Terminal)

MACROSCOPIC FINDINGS

Skin Scabs
Side/s of face: (Left , A few) 2mm

Skin Alopecia
Side/s of face

Lungs
Congested: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Nasal Passages
Degeneration of olfactory epithelium: (Trace)

Larynx
Subepithelial inflammation - ventral: (Moderate)

Lungs
Pneumonitis: (Trace)

Heart
Myocarditis: (Trace)

Liver
Parenchymal inflammatory cells: (Minimal)

Skin
Scab: (Minimal , Focus)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 29M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Pharynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 30M (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Infra-auricular region/s: (Minimal)

Lymph Nodes - Cervical
Congested: (Right)

Stomach Antrum Mucosa
White nodules, near to limiting ridge: (Two , Punctate)

Kidneys
Increased pelvic dilatation: (Right , Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Subepithelial inflammation - ventral: (Minimal)

Heart
Valvular cyst

Lymph Nodes - Cervical
Sinus histiocytosis: (Minimal)

Kidneys
Tubular basophilia: (Trace)
Dilatation of the renal pelvis: (Minimal , Unilateral)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 30M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Thymus; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 31M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested: (Minimal)

Stomach Antrum Mucosa
White nodules, near to limiting ridge: (Three) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal , Area , One lobe)
Perivascular inflammatory infiltration: (Trace)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 32M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 33M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few , Punctate)

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

Liver
Median cleft, pale subcapsular area/s: (One) 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 34M (Recovery)

MACROSCOPIC FINDINGS

Skin Scabs
Side/s of face: (Left , One) 2mm

Skin Alopecia
Side/s of face

Lungs
Congested

Kidneys
Increased pelvic dilatation: (Right , Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)
Vascular congestion

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Kidneys
Dilatation of the renal pelvis: (Minimal , Unilateral)

Skin
Epidermal hyperplasia: (Minimal , Area)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 34M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 35M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (Multiple , Punctate)

Liver
Median cleft, pale subcapsular area/s: (One) 2mm

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)
Aggregates of macrophages: (Trace)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 36M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few , Punctate)

Lymph Nodes - Tracheobronchial
Enlarged: 4mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 37M (Recovery)

MACROSCOPIC FINDINGS

Kidneys
Increased pelvic dilatation: (Right , Moderate)
Pelvis contained cloudy fluid: (Right)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal , One lobe)

Kidneys
Dilatation of the renal pelvis: (Minimal , Unilateral)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 38M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested
Pale subpleural foci: (Multiple , Punctate)

Lymph Nodes - Tracheobronchial
Enlarged: 4mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Moderate)
Perivascular inflammatory infiltration: (Moderate)
Prominent BALT: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 39M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested: (Minimal , Patchy)

Lymph Nodes - Tracheobronchial
Enlarged: 6x3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Minimal)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 40M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 41M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Subpleural aggregation of alveolar macrophages: (Minimal)
Aggregates of macrophages: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 42M (Terminal)

MACROSCOPIC FINDINGS

Liver
Median cleft, pale subcapsular area/s: (One) 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Liver
Vacuolated hepatocytes at median cleft

The following tissues were considered normal:

Lungs; Lymph Nodes - Mediastinal

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 43M (Terminal)

MACROSCOPIC FINDINGS

Forestomach
Cyst - limiting ridge: 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Stomach
Cysts at limiting ridge

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 44M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 45M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 46M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 47M (Terminal)

MACROSCOPIC FINDINGS

Forestomach
Cyst - limiting ridge: 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Stomach
Cysts at limiting ridge

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 48M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Subpleural aggregation of alveolar macrophages: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Low
Rat No/Sex:	49M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Aggregates of macrophages: (Trace , Focus)

Lymph Nodes - Mediastinal
Siderocytes: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 50M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	51M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial

Tissues not available for examination were:

Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 52M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	53M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)
Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 54M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial

Tissues not available for examination were:

Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	55M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 56M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	57M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 58M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Trace)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	59M (Terminal)

MACROSCOPIC FINDINGS

Epididymides
Yellow swelling/s: (Left , One) 4mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Testes
Atrophy: (Minimal)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Epididymides : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	60M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 61M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Moderate)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Mural hypertrophy of bronchial arteries
Brown pigment in macrophages: (Minimal)
Macrophages containing fine brown pigment in BALT: (Trace)

Heart
Myocarditis: (Trace)
Valvular endocarditis: (Minimal)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Minimal)

Lymph Nodes - Mediastinal
Macrophages containing brown pigment: (Minimal)

Testes
Atrophy: (Trace , Unilateral)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 61M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 62M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)
Macrophages containing fine brown pigment in BALT: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Trace)

Lymph Nodes - Mediastinal
Macrophages containing brown pigment: (Trace)

Urinary Bladder
Urothelial hyperplasia: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Liver; Pancreas; Kidneys; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 63M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Moderate)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Brown pigment in macrophages: (Trace)

Heart
Myocarditis: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Minimal)

Kidneys
Tubular basophilia: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 64M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Extension of bronchiolar epithelium into alveolar ducts: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)
Macrophages containing fine brown pigment in BALT: (Trace)

Heart
Myocarditis: (Trace)

Lymph Nodes - Mediastinal
Macrophages containing brown pigment: (Minimal)

Kidneys
Tubular basophilia: (Trace)
Medullary cyst

Sciatic Nerve
Degenerate fibres: (Trace)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 64M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 65M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Trace)
Increased collagen in alveolar duct walls: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Brown pigment in macrophages: (Trace)

Heart
Myocarditis: (Minimal)

Kidneys
Tubular basophilia: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 66M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)
Macrophages around alveolar ducts: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Mediastinal
Siderocytes: (Minimal)

Thyroids
Ectopic thymic tissue

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 67M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Minimal)
Extension of bronchiolar epithelium into alveolar ducts: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

Heart
Myocarditis: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Minimal)

Kidneys
Tubular basophilia: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	68M (Terminal)

MACROSCOPIC FINDINGS

Liver
Pale subcapsular area/s: (One) Median lobe 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Minimal)
Increased collagen in alveolar duct walls: (Trace)
Extension of bronchiolar epithelium into alveolar ducts: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)
Macrophages containing fine brown pigment in BALT: (Trace)

Heart
Myocarditis: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Minimal)

Lymph Nodes - Mediastinal
Macrophages containing brown pigment: (Minimal)

Liver
Vacuolated hepatocytes at median cleft: (Area)

Kidneys
Tubular basophilia: (Trace)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 68M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 69M (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Macrophages containing fine brown pigment in BALT: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Moderate)

Liver
Hepatocyte necrosis: (Minimal , Focus)

Kidneys
Tubular basophilia: (Minimal)
Cortical inflammatory cells: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Pancreas; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 70M (Terminal)

MACROSCOPIC FINDINGS

Stomach Corpus Mucosa
White nodule, near to limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)
Macrophages around alveolar ducts: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Brown pigment in macrophages: (Minimal)

Lymph Nodes - Cervical
Sinus erythrocytosis: (Minimal)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Minimal)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Epididymides; Testes; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 71M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few) 1mm

Lymph Nodes - Tracheobronchial
Enlarged: 5mm

Forestomach
Cyst - limiting ridge: 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)
Brown pigment in macrophages: (Minimal)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Stomach
Cysts at limiting ridge: (Moderate)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 72M (Recovery)

MACROSCOPIC FINDINGS

Lymph Nodes - Tracheobronchial
Enlarged: 6mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	73M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested

Lymph Nodes - Tracheobronchial
Enlarged: 5mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 74M (Recovery)

MACROSCOPIC FINDINGS

Stomach Antrum Mucosa
White nodules, near to limiting ridge: (Two) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 75M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 76M (Recovery)

MACROSCOPIC FINDINGS

Skin Scabs
Tail: (A few) 2mm

Lymph Nodes - Tracheobronchial
Enlarged: 8mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)
Prominent BALT: (Moderate)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Moderate)

Tail
Pustule

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 77M (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested

Epididymides
Pale area: (Left) 3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Epididymides : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 78M (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cervical region/s: (Left , Right)

Lungs
Pale subpleural foci: (A few) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Granulomatous inflammation: (Trace)
Perivascular inflammatory infiltration: (Moderate)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	79M (Recovery)

MACROSCOPIC FINDINGS

Skin Scabs
Sacral region: (A few) 3mm

Skin Alopecia
Sacral region

Lungs
Congested
Pale subpleural foci: (Multiple) 1mm

Lymph Nodes - Tracheobronchial
Enlarged: 6x3mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Skin
Scab: (Minimal)
Epidermal erosion: (Moderate)
Epidermal hyperplasia: (Minimal)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 79M - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	80M (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace)
Brown pigment in macrophages: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 81F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 82F (Terminal)

MACROSCOPIC FINDINGS

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Uterus
Luminal dilatation: (Minimal)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 83F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Tissues not available for examination were:

Lymph Nodes - Mediastinal : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 84F (Intercurrent)

MACROSCOPIC FINDINGS

Found dead

Lungs
Congested

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Vascular congestion: (Minimal)

Kidneys
Dystrophic mineralisation: (Trace)

Factors Contributory To Death
Unknown

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 85F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 86F (Terminal)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Subpleural aggregation of alveolar macrophages: (Trace)

Kidneys
Dystrophic mineralisation: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 87F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Heart
Myocardial fibrosis: (Minimal , Focus)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 88F (Terminal)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few) 1mm

Forestomach
Cyst - limiting ridge: 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Subpleural aggregation of alveolar macrophages: (Trace)

Heart
Myocarditis: (Trace , Focus)

Lymph Nodes - Mediastinal
Sinus congestion with siderocytes: (Minimal)

Stomach
Cysts at limiting ridge: (Moderate)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 89F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Interscapular region/s: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 90F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 91F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace , Some lobes)
Perivascular inflammatory infiltration: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 92F (Recovery)

MACROSCOPIC FINDINGS

Lymph Nodes - Mediastinal
Congested

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lymph Nodes - Mediastinal
Sinus congestion

The following tissues were considered normal:

Pharynx; Lungs; Lymph Nodes - Tracheobronchial

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 93F (Recovery)

MACROSCOPIC FINDINGS

Uterus
Fluid distension

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 94F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace , Focus , One lobe)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 95F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 96F (Recovery)

MACROSCOPIC FINDINGS

Skin Scabs
Tail: (One) 3mm

Tail
Pustule/s: (One , Scabby) 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Tail
Pustule
Ulceration: (Minimal)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 97F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 98F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Infra-auricular region/s

Subcutis
Mass - right inguinal region: A: 23x14x9mm

Pituitary
Swollen

Lungs
Pale subpleural foci: (A few , Punctate)

Lymph Nodes - Tracheobronchial
Enlarged: 3mm

Liver
Median cleft, pale subcapsular area/s: (One) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Trace)

Pituitary
Hyperplasia in the pars anterior: (Moderate)

Mammary Glands
Mammary adenocarcinoma: (NEOPLASTIC , MALIGNANT , PRIMARY) (A)

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 98F - continued

MICROSCOPIC FINDINGS - continued

The following tissues were considered normal:

Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 99F (Recovery)

MACROSCOPIC FINDINGS

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Air

Dosage Level: Control

Rat No/Sex: 100F (Recovery)

MACROSCOPIC FINDINGS

Uterus
Fluid distension

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 101F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Tissues not available for examination were:

Parathyroids : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 102F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Scapular region: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)
Subepithelial inflammation - ventral: (Trace)

Lungs
Subpleural aggregation of alveolar macrophages: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 103F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 104F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Trace)
Subepithelial inflammation - ventral: (Trace)

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

Kidneys
Dilatation of the renal pelvis: (Trace , Unilateral)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 104F - continued

MICROSCOPIC FINDINGS - continued

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 105F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Scapular regions: (Minimal , Patchy)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)

Lymph Nodes - Tracheobronchial
Macrophages containing haemosiderin: (Moderate)

Lymph Nodes - Mediastinal
Sinus congestion with siderocytes: (Minimal)

Liver
Parenchymal inflammatory cells: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 106F (Intercurrent)

MACROSCOPIC FINDINGS

Found dead

Incisors
Pale: (Lower)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Moderate)

Lungs
Vascular congestion: (Minimal)

Factors Contributory To Death
Unknown

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 107F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 108F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)

Lungs
Aggregates of macrophages: (Trace , Focal , One lobe)

Kidneys
Dystrophic mineralisation: (Minimal)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 108F - continued

MICROSCOPIC FINDINGS - continued

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 109F (Terminal)

MACROSCOPIC FINDINGS

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Minimal)
Subepithelial inflammation - ventral: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach : (W.N.L.); Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 110F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Larynx
Epithelial hyperplasia - ventral: (Moderate)
Subepithelial inflammation - ventral: (Minimal)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Trachea(including Bifurcation); Lungs; Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 111F (Recovery)

MACROSCOPIC FINDINGS

Kidneys
Increased pelvic dilatation: (Right , Moderate)
Irregular cortical scarring: (Right , Severe)

Ureters
Distended: (Right) 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Kidneys
Pyelonephritis: (Moderate , Unilateral)

The following tissues were considered normal:

Lungs; Lymph Nodes - Mediastinal; Ureters : (W.N.L.)

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 112F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Thoracic region/s: (Right)

Pituitary
Swollen

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal; Pituitary : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 113F (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested
Pale subpleural foci: (A few , Punctate)

Lymph Nodes - Tracheobronchial
Enlarged: 4mm

Uterus
Fluid distension

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 114F (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Minimal)
Perivascular inflammatory infiltration: (Moderate)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 115F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Thoracic region/s: (Right)
Abdominal region

Lungs
Congested
Pale subpleural foci: (A few , Punctate)

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Moderate)
Vascular congestion: (Minimal)

Uterus
Luminal dilatation: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 116F (Recovery)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 117F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 118F (Recovery)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few , Punctate)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 119F (Recovery)

MACROSCOPIC FINDINGS

Uterus
Fluid distension

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: NaF

Rat No/Sex: 120F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 121F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 122F (Terminal)

MACROSCOPIC FINDINGS

Forestomach
Cyst - limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Stomach
Cysts at limiting ridge: (Moderate)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Low
Rat No/Sex:	123F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 124F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lymph Nodes - Mediastinal
Siderocytes: (Trace)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 125F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 126F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Hindlimb/s: (Minimal)
Sacral region

Lymph Nodes - Cervical
Congested: (Minimal)

Lungs
Pale subpleural foci: (A few , Punctate)

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lymph Nodes - Cervical
Sinus erythrocytosis: (Minimal)

The following tissues were considered normal:

Lungs : (W.N.L.); Lymph Nodes - Mediastinal; Stomach : (W.N.L.)

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 127F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Periorbital region/s: (Minimal)

Adipose Tissue
Minimal

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 128F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Periorbital region/s: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 129F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Low

Rat No/Sex: 130F (Terminal)

MACROSCOPIC FINDINGS

Fur
Coarse - dorsum: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 131F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Ectopic bone focus

Lymph Nodes - Mediastinal
Siderocytes: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 132F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	133F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 134F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lymph Nodes - Mediastinal
Siderocytes: (Trace)

The following tissues were considered normal:

Lungs

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 135F (Terminal)

MACROSCOPIC FINDINGS

Liver
Median cleft, pale subcapsular area/s: (One) 1mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 136F (Terminal)

MACROSCOPIC FINDINGS

Lungs
Pale subpleural foci: (A few , Punctate) right posterior lobe

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 137F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 138F (Terminal)

MACROSCOPIC FINDINGS

Skin Alopecia
Periorbital region/s: (Right)

Liver
Congested lobe/s: posterior caudate
Firm lobe/s: posterior caudate

Uterus
Fluid distension

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Vascular congestion: (Minimal)

Liver
Hepatocyte necrosis: (Marked , One lobe)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	Intermediate
Rat No/Sex:	139F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following tissues were considered normal:

Lungs; Lymph Nodes - Mediastinal

Tissues not available for examination were:

Lymph Nodes - Tracheobronchial : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: Intermediate

Rat No/Sex: 140F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 141F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Macrophages containing fine brown pigment in BALT: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Trace , Focal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 142F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	143F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Trace)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Trace)

Lymph Nodes - Mediastinal
Sinus congestion with siderocytes: (Minimal)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 144F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Mediastinal
Macrophages containing brown pigment: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	145F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Minimal)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Minimal)

Lymph Nodes - Mediastinal
Sinus congestion with siderocytes: (Minimal)
Macrophages containing brown pigment: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 146F (Terminal)

MACROSCOPIC FINDINGS

Stomach Antrum Mucosa
White nodule, near to limiting ridge: 1mm

Kidneys
Increased pelvic dilatation: (Left , Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Trace)

Kidneys
Dilatation of the renal pelvis: (Minimal , Unilateral)

Stomach
Focus of ectopic non-glandular epithelium within the glandular mucosa

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

APPENDIX 18

(Pathology - continued)

Rat No/Sex: 146F - continued

MICROSCOPIC FINDINGS - continued

Tissues not available for examination were:

Parathyroids : (Not seen)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 147F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Trace)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

Liver
Parenchymal inflammatory cells: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 148F (Terminal)

MACROSCOPIC FINDINGS

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Moderate)

Kidneys
Dystrophic mineralisation: (Minimal)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Urinary Bladder; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 149F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

Heart
Myocarditis: (Trace)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Spleen; Liver; Pancreas; Kidneys; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 150F (Terminal)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Macrophages around alveolar ducts: (Minimal , Focal , One lobe)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Macrophages containing brown pigment: (Trace)

Lymph Nodes - Mediastinal
Macrophages containing brown pigment: (Minimal)

Kidneys
Dystrophic mineralisation: (Minimal)

The following tissues were considered normal:

Nasal Passages; Pharynx; Larynx; Trachea(including Bifurcation); Aorta; Heart; Thymus; Lymph Nodes - Cervical; Lymph Nodes - Mesenteric; Spleen; Liver; Pancreas; Urinary Bladder; Uterus; Cervix; Ovaries; Thyroids; Parathyroids; Adrenals; Pituitary; Salivary Glands; Oesophagus; Stomach; Duodenum; Jejunum; Ileum; Caecum; Colon; Rectum; Eyes; Sciatic Nerve; Brain; Sternum

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 152F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Brown pigment in macrophages: (Trace)

Lymph Nodes - Mediastinal
Siderocytes: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 153F (Recovery)

MACROSCOPIC FINDINGS

Subcutis
Mass - ventral cervical region: (Right) A: 32x28x13mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)
Brown pigment in macrophages: (Trace)

Mammary Glands
Mammary adenocarcinoma: (NEOPLASTIC , BENIGN , PRIMARY) (A)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 154F (Recovery)

MACROSCOPIC FINDINGS

No abnormalities detected

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)
Alveolitis with interstitial thickening of alveolar duct walls: (Trace)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 155F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Scapular region: (Diffuse)

Liver
Median cleft, pale subcapsular area/s: (One) 3mm

Uterus
Fluid distension: (Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Trace , Focal , One lobe)

Uterus
Luminal dilatation: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 156F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cranial region: (Minimal)

Lymph Nodes - Tracheobronchial
Enlarged: 7mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace)
Perivascular inflammatory infiltration: (Minimal)
Brown pigment in macrophages: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

The following tissues were considered normal:

Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	157F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cranial region: (Minimal)

Kidneys
Increased pelvic dilatation: (Left , Minimal)

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

Kidneys
Dilatation of the renal pelvis: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 158F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cranial region

Lungs
Congested: (Minimal)
Pale subpleural foci: (A few , Punctate)

Liver
Median cleft, pale subcapsular area/s: (One) 2mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Trace)
Perivascular inflammatory infiltration: (Trace)

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal; Liver : (W.N.L.)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound:	Cryolite
Dosage Level:	High
Rat No/Sex:	159F (Recovery)

MACROSCOPIC FINDINGS

Skin Alopecia
Cranial region
Cervical region/s: (Dorsal)

Lungs
Congested

Lymph Nodes - Tracheobronchial
Enlarged: 4mm

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Pneumonitis: (Moderate)
Perivascular inflammatory infiltration: (Trace)

Lymph Nodes - Tracheobronchial
Increased lymphoid cellularity - generalised: (Minimal)

Lymph Nodes - Mediastinal
Increased lymphoid cellularity - generalised: (Minimal)

Pathologist: R.L.Gregson

APPENDIX 18

(Pathology - continued)

Compound: Cryolite

Dosage Level: High

Rat No/Sex: 160F (Recovery)

MACROSCOPIC FINDINGS

Lungs
Congested
Pale subpleural foci: (A few , Punctate)

Ovaries
No corpora lutea visible

All the other organs and tissues appeared normal.

MICROSCOPIC FINDINGS

The following observations were noted:

Lungs
Perivascular inflammatory infiltration: (Minimal)

Ovaries
Absence of corpora lutea

The following tissues were considered normal:

Lymph Nodes - Tracheobronchial; Lymph Nodes - Mediastinal

Pathologist: R.L.Gregson

APPENDIX 19

Historical control data haematology parameters

Male CD (UK) Sprague Dawley Rats (19 Weeks of Age)

Parameter	Confidence Interval		
	5%	50%	95%
PCV	43	46	50
Haemoglobin	14.8	16.0	16.9
RBC	8.2	8.9	9.7
MCHC	32.7	34.4	35.9
MCV	49	52	55
MCH	17.0	18.0	19.5
WBC (total)	9.76	13.69	19.17
Neutrophils	0.91	1.64	3.30
Lymphocytes	7.86	11.38	15.19
Eosinophils	0.07	0.15	0.3
Basophils	0.02	0.04	0.07
Monocytes	0.10	0.24	0.49
Large unstained cells	0.06	0.27	0.68
Platelets	828	1053	1257
Retic App + Tab	1.0	2.1	3.8

APPENDIX 19

(Historical control data haematology parameters - continued)

Female CD (UK) Sprague Dawley Rats (19 Weeks of Age)

Parameter	Confidence Interval		
	5%	50%	95%
PCV	42	45	48
Haemoglobin	14.6	15.6	16.4
RBC	7.6	8.3	9.0
MCHC	33.2	34.8	36.2
MCV	51	54	57
MCH	17.8	18.9	20.0
WBC (total)	5.27	8.53	13.06
Neutrophils	0.50	0.87	2.17
Lymphocytes	3.99	6.79	11.15
Eosinophils	0.04	0.10	0.22
Basophils	0.01	0.02	0.05
Monocytes	0.06	0.14	0.31
Large unstained cells	0.05	0.13	0.33
Platelets	792	1035	1273
Retic App + Tab	0.80	1.40	3.30

SODIUM HEXAFLUOROALUMINATE (Natriumaluminiumfluorid)
CAS No. 13775-53-6

B G No. 107

90 DAY REPEAT DOSE INHALATION STUDY IN RATS
(snout only exposure)

Sponsor

BG Chemie,
Kurfürsten-Anlage 62,
69115 Heidelberg,
GERMANY.

Sponsor's representative

Dr. G Martens

Research Laboratory

Huntingdon Life Sciences Ltd.,
P.O. Box 2,
Huntingdon,
Cambridgeshire,
PE18 6ES,
ENGLAND.

Report issued: 26th November 1997

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APPENDIX 19

Historical control data haematology parameters

Male CD (UK) Sprague Dawley Rats (19 Weeks of Age)

Parameter	Confidence Interval		
12	5%	50%	95%
PCV	43	46	50
Haemoglobin	14.8	16.0	16.9
RBC	8.2	8.9	9.7
MCHC	32.7	34.4	35.9
MCV	49	52	55
MCH	17.0	18.0	19.5
WBC (total)	9.76	13.69	19.17
Neutrophils	0.91	1.64	3.30
Lymphocytes	7.86	11.38	15.19
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APPENDIX 19

(Historical control data haematology parameters - continued)

Female CD (UK) Sprague Dawley Rats (19 Weeks of Age)

Parameter	Confidence Interval		
	5%	50%	95%
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Large unstained cells	0.05	0.13	0.33
Platelets	792	1035	1273
Retic App + Tab	0.80	1.40	3.30

APPENDIX 19

Historical control data biochemistry parameters

Male CD (UK) Sprague Dawley Rats (19 Weeks of Age)

Parameter	Confidence Interval		
	5%	50%	95%
Total Protein	6.2	6.8	7.4
Albumin	2.7	2.9	3.8
Globulin	3.2	3.8	4.3
BUN	10	13	18
AP	113	168	230
Bilirubin (total)	0.1	0.2	0.3
Creatinine	0.4	0.5	0.6
Na	142	144	147
K	3.1	3.6	4.1
Ca	5.2	5.5	5.7
Inorganic P	3.4	3.8	4.5
Cl	97	101	104
Cholesterol	48	69	105
Glucose	93	117	152
GPT	22	29	39
GOT	44	57	73
γGT	1	1	3
CPK	57	94	204

APPENDIX 19

(Historical control data biochemistry parameters - continued)

Male CD (UK) Sprague Dawley Rats (19 Weeks of Age)

Parameter	Confidence Interval		
	5%	50%	95%
Total Protein	6.2	6.9	7.7
Albumin	2.9	3.3	4.3
Globulin	2.7	3.6	4.2
BUN	11	15	23
AP	65	99	151
Bilirubin (total)	0.1	0.2	0.3
Creatinine	0.5	0.6	0.7
Na	141	144	146
K	2.9	3.3	4.0
Ca	5.1	5.5	5.8
Inorganic P	2.3	3.3	3.9
Cl	98	102	105
Cholesterol	53	81	112
Glucose	93	117	144
GPT	18	25	47
GOT	43	54	94
γGT	1	1	3
CPK	60	82	174

APPENDIX 20

Analytical certificate of Sodium fluoride

GLP FINAL REPORT

Identity and material balance

Bayer AG		Date	: 09. Jan. 1996
ZF-Forschungsdienste		Study No.	: A 95/0134/00 LEV
Building Q 18		Study Director	: Dr. Seelemann
D-51368 Leverkusen		Deputy Director	: Dr. Hamm

Test substance : Sodium fluoride

Customer : Dr. Paetz, BALK, LEV, Building D 8 Order no: 95/053

Chemical name	: Sodium fluoride		
Empirical formula	: NaF	Molecular mass	: 41.99 g/mole
CAS-no.	: 7681-49-4	Charge/Lot no.	: --
Sample no./Year	: 914130/1995	Sample reception date: 29.11.95	
Produktion plant	: AI-AS-P, R 14	Date of production : --	
Product no.	: 00 00002151	Endurance limit	: 29.11.99

Start Date	: 11.12.95
Termination Date	: 09.01.96

1. Methods and results

1.1 Verification of specification and further investigations of balancing
Specification date: --
Specification data: --

1.1.1 Test: Identity (IR)
SOP: D 0085601 DZA
Supervisor: Dr. Seelemann
Result: Identity complies

Description of the method:
Triturate a small aliquot of the sample with potassium bromide and press a pellet. Record a FT-IR spectrum.

1.1.2 Test: Fluoride content
SOP: 2011-0330001-92D
Supervisor: Dr. Neupert
Result: 44.9 %; 45.0 % fluoride

Description of the method:
The sample is dissolved in water. After addition of foreign salt and buffer TISAB at pH 5.5 the solution is titrated potentiometrically using an ion selective electrode and a standard addition process.
Standard compound: 1250ALK1001, 99.5 %

1.1.3 Test: Sodium content (AAS)
SOP: 2011-0267201-91D
Supervisor: Dr. Wicker
Result: 558000 mg/kg sodium

APPENDIX 20

(Analytical certificate of Sodium fluoride - continued)

Description of the method:
The sample is disolved in water and hydrochloric acid. The content is determined by AAS at 330.2 nm. The atomisation is done with air/acetylene.

1.2 Material balance

45.0 % fluoride
55.8 % sodium

100.8 % content sodium fluoride

2. Evaluation and comments

- The identification was done by interpretation of the FT-IR spectrum.

- The material balance is complete with respect to the state of the art.

3. Archiving of records
GLP archives, Bayer AG, ZF-D Zentrale Analytik, building O 13, D-51368 Leverkusen.

The testing schedule, raw data, final report, inspection reports and all other data relevant for a reexamination are archived.

4. Archiving of the sample
GLP samples archives, Bayer AG, ZF-D Zentrale Analytik, building O 13, D-51368 Leverkusen.

5. Declaration of the study director
The investigations have been performed corresponding to the OECD prinziples of Good Laboratory Practice (GLP) from 04.02.83 (published in Bundesanzeiger Nr.42a of 02.03.83) and the prinziples of Good Laboratory Practice (GLP) with respect to appendix 1 of the law for protection against hazardous materials (Chemikaliengesetz) from 25.07.1994 (published in Bundesgesetzblatt, Teil I of 29.07.1994).

Study director:	10.1.96	orig. sign.
	(Date)	Dr. Seelemann

Enclusures: Declaration of the Quality assurance

Translator:	24.1.96	
	(Date)	Dr. Königer

APPENDIX 20

(Analytical certificate of Sodium fluoride - continued)

Bayer AG
ZF-D Zentrale Analytik
Building O 13
D-51368 Leverkusen

Quality assuranze declaration to the final report

Study no. or order no., respectively : A 95/0134/00 LEV
Title of the GLP investigation : Sodium fluoride
Kind of study : Identity and material balance

This GLP investigation was reviewed by the quality assurance unit. The dates of inspection and the dates of reports to the head of the testing institution and the study director are listed as follows:

Inspection (date)	Report (date)
08.12.95	08.12.95
10.01.96	10.01.96

The results cited in the final report of this study are inspected on the basis of the actual SOP's/analytical methods. Concerning our best knowledge they correspond to the existing raw data.

Quality assurance: 10.01.96 (date) — orig. sign. Dr. Dittrich

Translator: [signature] (Date) — 24.1.96 Dr. Königer

APPENDIX 21

Analytical certificate of Sodium hexafluoroaluminate

G L P - A b s c h l u ß b e r i c h t Seite 1 von 4

ANALYTISCHE STOFFBILANZ

BAYER AG
ZF-D Zentrale Analytik
Gebäude O 13
5090 Leverkusen 1

Datum : 27.Jan.1993
Studiennummer: A 92/0098/01 LEV
Prüfleiter : Dr.Möhnle
Vertreter : Dr.Imbeck

Prüfsubstanz: Kryolith synth. leicht gemahlen

Auftraggeber: Dr.Paetz, BALK, LEV, Gebäude D 8 Auftragsnummer: 92/076

Chemische Bezeichnung : Trinatrium-Aluminium-Hexafluorid

Summenformel	: AlF_6*3Na	Molare Masse	: 209,9 g/mol
CAS-Nr.	: 13775-53-6	Charge/Partie-Nr.	: 2
Proben-Nr./Jahr	: 486448/1992	Datum Probennahme	: 10.09.92
Herstellbetrieb	: AC-P3	Herstelldatum	: 29.08.92
Produkt-Nr.	: 002209-00	Haltbarkeit bis	: 10.09.97

Beginn der Prüfung : 01.12.92
Ende der Prüfung : 27.01.93

1. Beschreibung der Methoden und Einzelergebnisse

1.1 Prüfungen auf Einhaltung der Spezifikation und weitere Untersuchungen zur Bilanzierung
Spezifikation vom : --
Spezifikationswerte : --

1.1.1 Prüfung : Schmelzaufschluß
Methoden-Nr.: 2011-0362801-92D
Betreuer : Dr.Möhnle
Ergebnis : --

1.1.2 Prüfung : Natrium
Methoden-Nr.: 2011-0106301-90D
Betreuer : Dr.Möhnle
Ergebnis : 32,8 % Natrium

1.1.3 Prüfung : Aluminium
Methoden-Nr.: 2011-0249101-91D
Betreuer : Dr.Möhnle
Ergebnis : 13,5 % Aluminium

APPENDIX 21

(Analytical certificate of Sodium hexafluoroaluminate - continued)

ANALYTISCHE STOFFBILANZ

BAYER AG	Datum : 27.Jan.1992
ZF-D Zentrale Analytik	Studiennummer: A 92/0098/01 LEV
Gebäude O 13	Prüfleiter : Dr.Möhnle
5090 Leverkusen 1	Vertreter : Dr.Imbeck

Prüfsubstanz: Kryolith synth. leicht gemahlen

Auftraggeber: Dr.Paetz, BALK, LEV, Gebäude D 8 Auftragsnummer: 92/076

1.1.4 Prüfung : Silicium
Methoden-Nr.: 2011-0299001-91D
Betreuer : Dr.Möhnle
Ergebnis : 0,2 % Silicium

1.1.5 Prüfung : Phosphor
Methoden-Nr.: 2011-0106001-90D
Betreuer : Dr.Möhnle
Ergebnis : 83 mg/kg Phosphor

1.1.6 Prüfung : Sulfat
Methoden-Nr.: 2011-0348401-92D
Betreuer : Dr.Mauss
Ergebnis : 78 mg/kg / 78 mg/kg Sulfat

1.1.7 Prüfung : Glühverlust
Methoden-Nr.: DIN 53568 (Juli 74)
Betreuer : Dr.v.d.Emden
Ergebnis : 0,2 %

1.1.8 Prüfung : Fluor
Methoden-Nr.: SOP 12.10 AC-F/367
Betreuer : Dr.Dorn
Ergebnis : 52,58 % / 52,60 % Fluor

1.2 Bilanzierung

Gemessen	Theorie	Differenz
32,8 % Natrium	32,9 % Natrium	- 0,1 %
13,5 % Aluminium	12,9 % Aluminium	+ 0,6 %
52,6 % Fluor	54,3 % Fluor	- 1,7 %
98,9 % Summe	100,1 % Summe	

APPENDIX 21

(Analytical certificate of Sodium hexafluoroaluminate - continued)

G L P - A b s c h l u ß b e r i c h t Seite 3 von 4

ANALYTISCHE STOFFBILANZ

BAYER AG ZF-D Zentrale Analytik Gebäude O 13 5090 Leverkusen 1	Datum : 27.Jan.1992 Studiennummer: A 92/0098/01 LEV Prüfleiter : Dr.Möhnle Vertreter : Dr.Imbeck

Prüfsubstanz: Kryolith synth. leicht gemahlen

Auftraggeber: Dr.Paetz, BALK, LEV, Gebäude D 8 Auftragsnummer: 92/076

2. **Bewertung und Kommentar**

- Die Werte für Natrium und Aluminium wurden aus dem Abschlußbericht Identität Studiennummer A 92/0098/00 LEV übernommen.
- Aufgrund des Überbefundes von Aluminium in Höhe von 0,6 % und dem Unterbefund von Fluor in Höhe von 1,7 % muß ein Anteil in der Prüfsubstanz als AlF_3 vorliegen. (Al:F = 1:3 = 0,6:1,8)
- Stoffbilanz ist nach Stand der Technik vollständig
- Rohdaten sind überprüft und werden archiviert

3. **Erklärung des Prüfleiters**

Die Untersuchungen wurden in Übereinstimmung mit den OECD-Grundsätzen der Guten Laborpraxis (GLP) vom 04.02.83 (veröffentlicht im Bundesanzeiger Nr.42a vom 02.03.83) und den Grundsätzen der Guten Laborpraxis (GLP) gemäß Anhang 1 des Gesetzes zum Schutz vor gefährlichen Stoffen (Chemikaliengesetz) vom 14.03.1990 (veröffentlicht im Bundesgesetzblatt, Teil I vom 22.03.1990) durchgeführt.

4. **Archivierung von Aufzeichnungen**

GLP-Archiv, Bayer AG, ZF-DZA Analytik Leverkusen/OAL, Gebäude O 13, 5090 Leverkusen 1

Prüfplan, Rohdatenblätter, Chromatogramme, Spektren, weitere für eine Nachprüfung relevante Unterlagen, Abschluß- und Inspektionsberichte.

5. **Archivierung des Rückstellmusters**

GLP-Probenlager, Bayer AG, ZF-DZA Analytik Leverkusen/OAL, Gebäude O 13, 5090 Leverkusen 1

Anlagen: Erklärung der Qualitätssicherungseinheit (QS)

APPENDIX 21

(Analytical certificate of Sodium hexafluoroaluminate - continued)

G L P - A b s c h l u ß b e r i c h t Seite 4 von 4

ANALYTISCHE STOFFBILANZ

BAYER AG ZF-D Zentrale Analytik Gebäude O 13 . 5090 Leverkusen 1	Datum : 27.Jan.1992 Studiennummer: A 92/0098/01 LEV Prüfleiter : Dr.Möhnle Vertreter : Dr.Imbeck

Prüfsubstanz: Kryolith synth. leicht gemahlen

Auftraggeber: Dr.Paetz, BALK, LEV, Gebäude D 8 Auftragsnummer: 92/076

	Prüfleiter:	01/02/93 (Datum)	
Betreuer:	(Dr.Mause)	2.2.93 (Datum)	
	(Dr.v.d.Emden)	2.II.93 (Datum)	
	(Dr.Dorn)	3.2.93 (Datum)	

Verteiler: Dr.Paetz, Bayer Altstoffkommission, LEV, Gebäude D 8
Archiv
Leiter Prüfeinrichtung
QS/ZF-DZA Koordination Alt- und Neustoffanalytik
Dr.Winkhaus, AC-P, LEV, Gebäude R 14
Dr.Hochgeschwender, AC-S, LEV, Gebäude O 1
Dr.Möhnle

APPENDIX 21

(Analytical certificate of Sodium hexafluoroaluminate - continued)

Bayer AG
ZF-DZA Analytik Leverkusen/OAL
Gebäude O 13
5090 Leverkusen 1

Anlage 1 zum
Abschlußbericht
Seite 1 von 1

Qualitätssicherungserklärung zum Abschlußbericht

Studien-Nr. bzw. Auftrag-Nr.: A 92/0098/01 LEV
Titel der GLP-Untersuchung : Kryolith synth. leicht gemahlen

Art der Studie: Charakterisierung/Stoffbilanz

Diese GLP-Untersuchung wurde laufend durch die Qualitätssicherung überprüft. Die Zeitpunkte der Inspektionen und die Zeitpunkte der Berichte an den Leiter der Prüfeinrichtung und an den Prüfleiter sind nachfolgend aufgeführt:

Überprüfung (Datum)	Bericht (Datum)
1. 12. 92	1. 12. 92
3. 3. 93	3. 3. 93

Die im Abschlußbericht dieser Untersuchung wiedergegebenen Ergebnisse werden auf der Basis der aktuellen SOP,'s/Analysenmethoden überprüft. Sie entsprechen nach unserem besten Wissen den vorliegenden Rohdaten.

Qualitätssicherung:

3.3.93
(Datum)

W. Willers
(WILLERS)

APPENDIX 22

Validation of method for the analysis of fluoride and aluminium contact of bone of teeth

SPONSOR

HUNTINGDON LIFE SCIENCES
(HLS)

STUDY SCHEDULE

HRC 1-8

STUDY OBJECTIVE

Validation of Method for the Analysis
of Fluoride and Aluminium Content
of Bone and Teeth

TESTING FACILITY

BUTTERWORTH LABORATORIES LIMITED
(BLL)

STUDY SCHEDULE NUMBER

BL 11/0366 (95)

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

STATEMENT OF DATA CONFIDENTIALITY

All data generated by Butterworth Laboratories Limited will be held in our archives for a period of not less than SIX years.

As with all work undertaken by Butterworth Laboratories Limited, for Huntingdon Life Sciences Limited, customer confidentiality is observed in accordance with our Confidentiality Agreement, signed by us on 4th January 1993.

STUDY DIRECTOR'S AUTHENTICATION

I, the undersigned, accept responsibility for the conduct of this study and confirm that analyses were undertaken in compliance with the Principles of Good Laboratory Practice.

Signed .....
Study Director

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

CONTENTS

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

PERSONNEL INVOLVED

DAVID A RICHES BSc, CChem, MRSC
Study Director

DAVID J HAWKINS BSc, C.Biol. M.I.Biol

DAVID W HUNT C.Chem MRSC

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

INTRODUCTION

Bone and tooth samples were received on 8 November 1995 in order to validate methods for the determination of Aluminium and Fluoride contents.

Fluoride was analysed using a method based on published methodology. Ashed, ground bone was dissolved in hydrochloric acid and the solution analysed by ion selective electrode.

Aluminium was analysed by fusing the bone in a sodium carbonate/boric acid mix. After neutralisation in hydrochloric acid, aluminium was detected in solution by Inductively Coupled Plasma Emission Spectrometry (ICP) at 167.08 nm.

EXPERIMENTAL PROCEDURE

Equipment/Instrumentation

Fluoride

- Kent Industrial Instruments 8001-2 Fluoride Ion Selective Electrode
- BDH Double Junction Glass Reference Electrode
- Ultralab 2100 pH/mv Meter
- Magnetic Stirrer and Stirring Fleas
- Plastic Beakers 25-40 ml capacity
- Mettler AT261 5 Figure Balance
- Grade A Volumetric Glassware
- Muffle Furnace set at 500°C

Aluminium

- Platinum Thimbles
- Plasma 300 ICP
- Plastic Volumetric Flasks

Reagents and Standards

- AnalaR grade Sodium Fluoride
- Total Ionic Strength Adjustment Buffer (TISAB)
- 0.25 N Hydrochloric Acid
- 0.125 N Sodium Hydroxide
- 1000 ppm Aluminium stock standard for AA - supplied by BDH
- AnalaR grade Sodium Carbonate
- AnalaR grade Boric Acid
- 1+1 Hydrochloric Acid

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

ANALYTICAL PROCEDURE

Calibration and analysis was carried out in accordance with Butterworth Laboratories Limited documented In-house method BLM 225. A copy is contained in the appendix of this report. Validation was performed by conducting a series of replicate analyses along with spiked samples.

SUMMARY OF RESULTS

Average weight of bone per animal	0.70g ashed
Average weight of teeth per animal	0.07g ashed

Minimum sample requirements for duplicate analysis is 100 mg ie the pooled bone and teeth of two animals will be required.

Analysis of composite sample for fluoride

Bone

Date 16th January to 18th January 1996	Result 0.036% ± 0.008
Date 13th February 1996	Result 0.041% ± 0.005

Spike recoveries obtained *76%, 98% and 98%.

* This was a spike at low concentration, where varations between samples runs have a more significant effect.

Teeth

Date 28th February 1996 Result 0.021% ± 0.002

Limit of detection 0.005%
Precision ± 0.005 or ± 10% of content whichever is greater

Analysis of composite sample for Aluminium

Bone

Date 22nd January 1996	Result 0.016% ± 0.001
Date 13th February 1996	Result 0.017% ± 0.004

Spike recoveries obtained 87% and 91%

Teeth

Date 26th February 1996 Result 0.010%
(Result obtained in duplicate)

Limit of detection 0.005%
Precision ± 0.005 or 10% of content whichever is greater.

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

INTERFERENCES

Samples of bone were spiked with possible interfering elements to determine their effect on the results.

Fluoride

The following elements were found to not produce a significant effect on the result

Aluminium at 0.08% in bone

Calcium at 20% in bone

Aluminium

Fluoride at 0.07% in bone

Phosphorus produces an emission signal close to Aluminium at 167 nm. However, a 1ppm aluminium standard was not effected by spiking with 250ppm phosphorus.

CONCLUSION

The results suggest that the methods used are suitable for extracting and analysing fluoride and aluminium in both tooth and bone samples.

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

PROTOCOL

SCHEDULE NO:

BL 11/0366 (95)

SPONSOR

Huntingdon Life Sciences Limited
PO Box 2, Huntingdon, Cambridgeshire PE18 6ES

TEST MATERIAL

8 samples of Rat Bone and Teeth
HRC Ref: 1-8
BL Ref: BL11/0366(95) - BL11/0373(95)

OBJECTIVE OF STUDY

Validation of Method for the Analysis of Fluoride and Aluminium Content of Bone and Teeth. Validation to be based on spiked recoveries and checking for cross-interference of the alternate ion, given the insolubility of the Aluminium Fluoride complex.

TESTING FACILITY

Butterworth Laboratories Limited
54-56 Waldegrave Road, Teddington, Middlesex TW11 8LG

STARTING DATE

December 1995.

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

COMPLETION DATE

January 1996.

A copy of the Certificate of Analysis will be issued as an interim report, with the original Certificate of Analysis being bound in the final GLP report to be issued within 25 working days of completion of the analytical work.

METHODS TO BE USED

Documented methods to be produced in Report following method validation.

Sample preparation methods to be compared will be Ash/Acid Dissolution and Ash/Fusion/Acid dissolution. Method of analysis proposed are: Ion Selective Electrode for Fluoride and Inductively Coupled Plasma Emission Spectrometry for Aluminium.

RECORDS TO BE ARCHIVED

All study specific raw data Instrumentation used, calibration standard results, all readings generated, all relevant sample/batch and reference numbers. Authorised copies of relevant raw data will be included in the final Report.

STUDY DIRECTOR

David A Riches BSc CChem MRSC
Senior Manager

MONITORING SCIENTIST

Derek Coombs
Division of Toxicology
Huntingdon Life Sciences Limited

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

ACCREDITATION

The Study will be conducted in compliance with the Principles of Good Laboratory Practice (GLP) as set forth in "Good Laboratory Practice, the United Kingdom Compliance Programme, Department of Health and Social Security, 1986, and subsequent revision, Department of Health 1989" and Good Laboratory Practice in the Testing of Chemicals OECD ISBN 92-64-12367-9, Paris 1982, subsequently republished OECD Environment Monograph No 45 1992.

Accepted on behalf of Butterworth Laboratories Limited

Signed..... Date....21/12/95....

David A Riches BSc CChem MRSC
Senior Manager
Study Director

Accepted on behalf of Huntingdon Life Sciences Limited

Signed..... Date..3 Jan 1995....

Derek Coombs
Division of Toxicology
Monitoring Scientist

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

BUTTERWORTH LABORATORIES LTD - IN-HOUSE METHOD

CONTROLLED DOCUMENT

METHOD NO: BLM 225

ISSUE NO: 1

DATE ISSUED: 29.5.96

ORIGINATOR: D Riches

AUTHORISED: T.Well

TITLE: The Determination of Aluminium and Fluoride in Tooth and Bone Samples

INTRODUCTION

Tooth and bone samples extracted post mortem are ashed at 500°C. Fluoride is determined in an acid extract of the ground material by Ion Selective Electrode (ISE). Aluminium is determined by fusing the ground material in a mix of sodium carbonate and boric acid. After acidification the solution is analysed for aluminium by inductively coupled plasma emission spectrometry (ICP).

LITERATURE REFERENCES

Determination of Fluoride in Bone with the Fluoride Electrode, Leon Singer and W D Armstrong

Standing Committee of Analysts "Fluoride in Waters, Effluents, Sludges, Plants and Soils 1982 (HMSO)

Decomposition Methods in Analytical Chemistry, Rudolf Bock, translated by Iain L Marr

METHOD VALIDATION

Validation was carried out on behalf of Huntingdon Life Sciences, BLL Study Schedule 11/0366 (95).

EQUIPMENT

Sample Preparation:	Silica Basin, Dessicator, Muffle Furnace @ 500°C, Small Pestle and Mortar, Waterbath
Fluoride:	Kent Ind. Inst. 8001-2 Fluoride ISE (or equivalent), Double Junction Glass Reference Electrode, pH/ISE meter, Magnetic Stirrer and Stirring Fleas, Plastic Beakers (25-40 ml), Analytical Balance (5-figure), Grade A Volumetric Glassware
Aluminium:	ICP, Platinum Thimbles, Plastic volumetric flask

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

BUTTERWORTH LABORATORIES LTD - IN-HOUSE METHOD

METHOD No: BLM 225 **ISSUE No: 1**

REAGENTS

Total Ionic Strength Adjustment Buffer (TISAB)
SpectrosoL or equivalent 1000 ppm Aluminium Stock Standard for AAS
AnalaR or equivalent: Sodium Carbonate (anhydrous), Boric Acid

1+1 Hydrochloric acid prepared from AnalaR or equivalent Hydrochloric Acid

0.25N Hydrochloric Acid, 0.125N Sodium Hydroxide prepared by dilution of Stock 1N AnalaR or equivalent solutions.

Primary Stock Fluoride Standard (1 ml ≡ 1000 μg F)

Dry AnalaR Sodium Fluoride for four hours at 105°C then cool in a dessicator. Accurately weigh 2.210 g ± 0.001 g and dissolve in water. Transfer to a one litre volumetric flask and make up to the mark. Immediately transfer to a polythene bottle. This solution is stable for three months.

INTERFERENCES

Fluoride: Hydroxyl ions which should be buffered against by using TISAB.

Chemical species that reduce the concentration of Fluoride ions in the sample, eg Polyvalent cations such as Calcium, Iron, Aluminium. The CDTA in the TISAB should decomplex combinations of Fluoride in these species.

Aluminium: A phosphorus emission signal occurs close to the Aluminium at 167.08 nm. Interference may occur by incorrect peak identification if Aluminium concentrations are low. A narrow peak search window is recommended to eliminate interferences.

SAMPLE PREPARATION

Place the bone or teeth sample into a silica basin and ash overnight at 500°C in a muffle furnace. After cooling in a dessicator, the material is ground to a fine powder in a pestle and mortar.

The minimum amount of ashed material required to perform a duplicate analysis for Aluminium and Fluoride is 100 μg.

The analysis for teeth and bone then follows the relevant analytical procedure.

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0346 (95)

BUTTERWORTH LABORATORIES LTD - IN-HOUSE METHOD

METHOD No: BLM 225 **ISSUE No: 1**

CONTROLLED DOCUMENT

ANALYTICAL PROCEDURE

Fluoride: Weigh accurately approximately 25 mg of ashed sample into a plastic 50 ml beaker. Add 5 ml of 0.25 N hydrochloric acid, and warm in a waterbath to dissolve. Cool, add 5 ml of 0.125 N sodium hydroxide and make up to 25 ml in a plastic volumetric flask.

Prepare a series of matrix matched Fluoride calibration standards by diluting the stock standard. Add equivalent quantitites of hydrochloric acid and sodium hydroxide as above before making to the mark.

Recommended standard concentrations are 0.01, 0.1, 0.5 and 5.0 mg/l.

Samples and standards are diluted 1 + 1 with TISAB prior to analysis by ISE in accordance with the following procedure:

Ion selective electrodes are sensitive instruments and various instability effects may be seen. Refer to the Standing Committee of Analysts "Fluoride in Waters, Effluents, Sludges, Plants and Soils 1982", section A8, page 14, before carrying out the experimental procedure.

(a) A constant temperature should be maintained throughout the run.

(b) Connect the ion selective electrode and the reference electrode to the pH meter and stand in TISAB overnight

(c) Switch on the pH meter and note the reading in mV. This is the background reading which must be achieved before a determination is made

(d) Prepare the working standards as detailed above

(e) Using a calibrated Gilson auto-pipette add 5ml of sample to a plastic beaker, followed by 5ml of TISAB and allow to stand for 15 minutes (check the pH - see note (k))

(f) Pour ≈ 10 ml of each working standard into plastic beakers and add a PTFE magnetic stirring flea to each beaker (sample and standards).

(g) Place the beaker onto the magnetic stirrer and establish a constant stirring rate so that no bubbles or vortex are formed. Fix the speed control on the stirrer to prevent the stirring rate from changing throughout the run

(h) Insert the electrode into the test solution, taking care not to trap any bubbles under the electrodes

(i) Leave for four minutes and switch off the magnetic stirrer (at the on/off switch). After one minute note the mV reading to nearest 1 mV. After a further minute note the reading again and if the readings are the same, record them. If not, repeat the process

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

BUTTERWORTH LABORATORIES LTD - IN-HOUSE METHOD

METHOD No: BLM 225 **ISSUE No: 1**

ANALYTICAL PROCEDURE Cont.

(j) Lift the electrodes out of the solutions and rinse thoroughly with deionised water. Dry with a clean tissue and immerse into a beaker containing TISAB. Allow the mV reading to reach the background level, rinse, dry and repeat from stage (f) onwards

(k) Analyse the samples in the same fashion as for the standards. The pH should be checked using pH strips. A pH in the range of 5-6 is desired. If this range is not achieved, a greater proportion of TISAB to sample may be used.

(l) A calibration graph is plotted:

y axis (on a log scale) = concentration in the test solution

x axis (on a linear scale) = - mV reading

From this graph (or from a statistical analysis carried out on a computer, based on an equation formed from the calibration data) the sample test solution concentrations can be calculated

Aluminium: Accurately weigh approximately 30 mg of ashed sample into a Platinum Thimble. Add 200 mg ± 20 mg sodium carbonate and 20 mg ± 5 boric acid to the thimble.

Carefully mix with a platinium wire, then heat over a bunsen flame until a clear melt is obtained.

Allow to cool, add 50% hydrochloric acid dropwise until the melt dissolves. Transfer the solution to a 25 ml plastic volumetric flask and make up to volume.

Prepare a series of matrix matched Aluminium calibration standards, including a blank, with equivalent amounts of sodium carbonate and boric acid as above. Recommended standard concentrations are 0.2, 0.5 and 1.0 ppm.

Aluminium is analysed by ICP at 167.08 nm in accordance with SOP IM 66.

$$\text{mg/kg Aluminium} = \frac{\text{mg/l Aluminium in solution} \times 25}{\text{sample weight in g}}$$

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

BUTTERWORTH LABORATORIES LTD - IN-HOUSE METHOD

METHOD No: BLM 225 **ISSUE No: 1**

PERFORMANCE CHARACTERISTICS

Accuracy	± 10% of content
Precision	± 10% of content
Limit of Detection	0.005%
Range	0.01 to 1.0% (higher ranges are achieved by dilution)

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

Page Ref. 0599

BUTTERWORTH LABORATORIES LIMITED
ANALYTICAL DATA RECORD SHEET

Name of Client: Huntingdon Life Sciences

BLL Reference(s): (check Customer Ref. etc.) 11/0366 – 11/0373 (95) Composite Bone

Methodology: Fluoride

Instrumentation: (Calibration Status etc.) ISE

Quality Control Employed: Replicate analysis. Spikes

Reference Materials: (If required) Analar Sodium Fluoride

Analytical Data:

To ~25mg of sample add 5ml 0.25N HCl. Dissolve in water bath. Add 5ml 0.125N NaOH and make up to 25ml in plastic volumetric flask. Dilute 1+1 with TISAB for analysis

Flask ID	Sample wt/mg	mg/l F	mg/kg in bone
1	23.0	0.31	340
2	38.7	0.57	370
3	23.7	0.23	350
4	54.2	0.36	(170) result anomaly. not used in calc.
5	11.6	0.17	370 *
6	32.1 + 10.8 µg F	0.79	76% recovery (assuming average 358 ppm)
7	29.3 + 21.7 µg F	1.27	98% recovery
8	27.0 + 10 µg Al	0.38	350
9	24.8 + 20 µg Al	0.38	380
10	23.8 + 2mg Ca	0.35	370
11	19.3 + 4 mg Ca	0.26	340

Analyst..... Date. 18/1/96

Checked by..... Date. 26/1/96

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0346 (95)

Huntingdon Life Sciences Validation of Fluoride in ne of Teeth by ISE

17/1/96 - Samples prepared by D.A. Riches
Ref:- P133

mple preparation diluted 50/50 in TISAB (BDH)
d allowed to stand for 5mins in plastic beakers.

d preparation:

NaF was dried overnight at 105°C and 1.1052 mg weighed out and dissolved in DI H_2O volume was made up to 1000mL to give a concentration of Fluoride in solution of:

499.97 mg/L

he working standards were prepared from this stock and an standard had equivalent volumes of HCl and NaOH had, to the sample preparation. See DAR B p
ach working standard was diluted 50/50 in TISAB and ved to stand for 5mins.

After this time each std and each sample was read in the F^- ISE following the procedure outlined in BLM +3 Section 2 (e) from and adding PTFE magnetic stirring flea...

Calibration and evaluation carried out using XY MATH.

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

225

17/1/96

Beaker /std.	mV	mg/L F^-
5 mg/L F^-	-142	Ø DSM 17/1/96
1 mg/L F^-	-102	
0.5 mg/L F^-	-87	
0.1 mg/L F^-	-54	
0.05 mg/L F^-	-39	
Blank	-36	0.04
1	-77	0.31
2	-90	0.57
3	-78	0.33
4	-80	0.36
5	-64	0.17
6	-97	0.79
7	-108	1.27
8	-81	0.38
9	-81	0.38
10	-79	0.35
11	-73	0.26

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

BUTTERWORTH LABORATORIES LIMITED
ANALYTICAL DATA RECORD SHEET

Name of Client: Huntingdon Life Sciences

BLL Reference(s): 11/0366(95) to 11/0373(95) Composite bone
(check Customer Ref. etc.)

Methodology: Aluminium by ICP. Na_2CO_3/H_3BO_3 fusion → 25 mls

Instrumentation: ICP
(Calibration Status etc.)

Quality Control Employed: Control standard run with samples

Reference Materials: 1000ppm Al standard ex BDH
(If required)

Analytical Data:

Dish No	Sample wt/mg	mg/l Al	% w/w Al
E	Blank	<0.02	
K	33.9	0.204	0.015
N	38.8	0.249	0.016
P	43.6	0.281	0.016

Analyst..... Date. 22/1/96

Checked by..... Date. 25/1/96

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P108: QWH book
DWH. 3. 100108 22/1/96

P 300 124660-00 22 JAN 96

SAMPLE	OPERATOR	PWR	TIME	NAME
RECAL	DR	3	11:05	AL-LOW

EL	NM	CH	SEC
AL	167.08	B	3.0

B#	INTENSITY	RAW-CONC	CONC
99	21	0.0006	0.000
1	125423	1.000	0.999

0 -62 x 61 x2

EL	NM	C	INTENSITY	CONC
AL	167.08	B	2130	0.015
AL	167.08	B	1401	0.010
AL	167.08	B	1545	0.011

P 300 124660-00 22 JAN 96

SAMPLE	OPERATOR	PWR	TIME	NAME
1	DR	3	11:07	AL-LOW

E

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	0.012	PPM	0.0026	21.66	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	27026	0.204
AL	167.08	B	27090	0.204
AL	167.08	B	27118	0.205

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

2	DR		3	11:08	AL-LOW			≡ 0.015%

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	0.204	PPM	0.0007	0.34	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	32884	0.249
AL	167.08	B	32974	0.249
AL	167.08	B	33134	0.251

P 300 124660-00 22 JAN 96

SAMPLE	OPERATOR	PWR	TIME	NAME	
3	DR	3	11:10	AL-LOW	N

35.5 mg/25 ml

EL	NM	CH	CONC	UNIT	SD	RSD	SEC	
AL	167.08	B	0.249	PPM	0.0012	0.48	3.0	≡ 0.016 %

EL	NM	C	INTENSITY	CONC
AL	167.08	B	36834	0.279
AL	167.08	B	37186	0.282
AL	167.08	B	37209	0.282

P 300 124660-00 22 JAN 96

SAMPLE	OPERATOR	PWR	TIME	NAME	
4	DR	3	11:11	AL-LOW	P

45.6 mg/25 ml

EL	NM	CH	CONC	UNIT	SD	RSD	SEC	
AL	167.08	B	0.281	PPM	0.0017	0.60	3.0	≡ 0.016 %

EL	NM	C	INTENSITY	CONC
AL	167.08	B	53471	0.410
AL	167.08	B	53671	0.411
AL	167.08	B	53278	0.408

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

P 300 124660-00 22 JAN 96

SAMPLE	OPERATOR	PWR	TIME	NAME
400	DR	3	11:13	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	0.409	PPM	0.0015	0.36	3.0

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

22/1/96

[redacted]

1/ 0511 Zn TMB 0.4995 → 10 2.50 mg/L = 50
C 0.5006 → 10 2.82 mg/L = 56

Pd 1 0.4999 → 5 ≤0.1 mg/L = <2 mg/kg
2 0.4911 → 5 <0.1 mg/L = <2 mg/kg

HLS-BL 11/0366(95)

Al in BONE. - MICRO Pt crucs. fused over Meker 0.2g ANH. Na_2CO_3/0.02g H_3BO_3
leached (1+1) Aerst-HCl (in conc) → 25 ml

1) E BL 0.012 mg/L

2) K 33.9 mg 0.204 0.014 %
3) N 38.8 mg 0.249 0.015 %
4) P 43.6 mg 0.281 0.015 %

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

Page Ref. 0604

BUTTERWORTH LABORATORIES LIMITED
ANALYTICAL DATA RECORD SHEET

Name of Client: Huntingdon Life Sciences

BLL Reference(s): (check Customer Ref. etc.) 11/0366 – 11/0373(95) Composite sample of bone

Methodology: Aluminium by ICP

Instrumentation: (Calibration Status etc.) ICP

Quality Control Employed: Control standards and well samples. Spikes

Reference Materials: (If required) 1000 ppm Al standard ex BDH

Analytical Data:

Method. To ~30 mg in a Pt thimble, add 0.2g Na_2CO_3 and 0.02g H_3BO_3. Fuse over burner. Cool. Add drops 50% HCl to dissolve melt. Dilute to 25 ml in plastic volumetric

Sample No	Sample wt/mg	mg/l Al	% w/w Al in bone
1	18.60	0.13	0.017
2	28.97	0.19	0.016
3	32.47	0.22	0.017
4	50.35	0.26	0.013
5	24.32	0.19	0.020
6	Blank	<0.02	
8	28.31 + 20µg Al	0.88	87% recovery (assuming 0.0166% Al in bone)
9	28.68 + 20µg Al	0.92	91% recovery
10	30.29 + 21.6µg F	0.17	0.014
11	34.42	0.24	0.017
12	29.45	0.27	(0.023 %) outlier result not included in calculation
13	Blank	<0.02	

Phosphorus produces an emission signal close to Al 167. However 1 ppm Al standard was not effected by a 250 ppm Phosphorus spike.

Analyst..... Date 13/2/96

Checked by..... Date 16/2/96.

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Effect of Phosphorus on Aluminium @ 167.0nm

EL	NM	CH	SEC
P	213.63	A	1.0

B#	INTENSITY	RAW-CONC	CONC
99	119	0d	0
10	8124	250	250

0 0 x 0 x2

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
RECAL	GH	3	07:14	AL-LOW

EL	NM	CH	SEC
P	213.63	A	1.0

B#	INTENSITY	RAW-CONC	CONC
99	94	0d	0
10	7981	250	250

0 0 x 0 x2

EL	NM	CH	SEC
AL	167.08	B	3.0

B#	INTENSITY	RAW-CONC	CONC
99	1341	0.000	0.000
1	72006	1.000	0.999

0 -62 x 61 x2

EL	NM	C	INTENSITY	CONC
P	213.63	A	64	-1
AL	167.08	B	69352	0.958
P	213.63	A	56	-2
AL	167.08	B	70368	0.973
P	213.63	A	61	-2
AL	167.08	B	71100	0.985

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
1	GH	3	07:16	AL-LOW

1ppm Al

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	-2d	PPM	0.7	35.00	1.0
AL	167.08	B	0.972	PPM	0.0135	1.38	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	7934	248
AL	167.08	B	70315	0.973
P	213.63	A	7983	250
AL	167.08	B	70371	0.973
P	213.63	A	8047	252
AL	167.08	B	70124	0.970

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
2	GH	3	07:18	AL-LOW

250ppm P
0.5ppm Al

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	250	PPM	2.0	0.80	1.0
AL	167.08	B	0.972	PPM	0.0017	0.17	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	91	-1
AL	167.08	B	35769	0.470
P	213.63	A	78	-1
AL	167.08	B	35020	0.460
P	213.63	A	71	-1
AL	167.08	B	33681	0.441

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
4	GH	3	07:20	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	-1d	PPM	0.0	0.00	1.0
AL	167.08	B	0.457	PPM	0.0147	3.21	3.0

EL	NM	CH	SEC
AL	167.08	B	3.0

B#	INTENSITY	RAW-CONC	CONC
99	1164	0.000	0.000
1	68651	1.000	0.999
4	28331	0.402	0.399

0 -9 x B x2

EL	NM	C	INTENSITY	CONC
P	213.63	A	4176	129
AL	167.08	B	10630	0.138
P	213.63	A	4011	124
AL	167.08	B	10240	0.132
P	213.63	A	3964	122
AL	167.08	B	10368	0.134

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
1	GH	3	07:25	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	125	PPM	3.6	2.88	1.0
AL	167.08	B	0.134	PPM	0.0030	2.23	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	6352	198
AL	167.08	B	14321	0.192
P	213.63	A	6207	193
AL	167.08	B	14478	0.195
P	213.63	A	6141	191
AL	167.08	B	14449	0.194

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
2	GH	3	07:27	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	194	PPM	3.6	1.85	1.0
AL	167.08	B	0.193	PPM	0.0015	0.77	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	7185	224
AL	167.08	B	19406	0.268
P	213.63	A	7173	224
AL	167.08	B	19220	0.265
P	213.63	A	7192	224
AL	167.08	B	19333	0.267

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
3	GH	3	07:29	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	224	PPM	0.0	0.00	1.0
AL	167.08	B	0.266	PPM	0.0015	0.56	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	11020	346
AL	167.08	B	18740	0.258
P	213.63	A	10901	342
AL	167.08	B	18651	0.257
P	213.63	A	10057	315
AL	167.08	B	18489	0.254

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
4	GH	3	07:30	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	334x	PPM	16.8	5.02	1.0
AL	167.08	B	0.256	PPM	0.0021	0.82	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	55	-2
AL	167.08	B	28103	0.396
P	213.63	A	46	-2
AL	167.08	B	28473	0.401
P	213.63	A	55	-2
AL	167.08	B	28361	0.399

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
0.4pp-Al 0	GH	3	07:32	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	-2d	PPM	0.0	0.00	1.0
AL	167.08	B	0.398	PPM	0.0025	0.62	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	5475	170
AL	167.08	B	14158	0.190
P	213.63	A	5341	166
AL	167.08	B	14037	0.188
P	213.63	A	5118	159
AL	167.08	B	13955	0.187

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
5 ~~41~~	GH	3	07:34	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	165	PPM	5.5	3.33	1.0
AL	167.08	B	0.188	PPM	0.0015	0.79	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	29	-3
AL	167.08	B	2525	0.019
P	213.63	A	44	-2
AL	167.08	B	2568	0.019
P	213.63	A	36	-2
AL	167.08	B	2509	0.018

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
6 ~~42~~	GH	3	07:35	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	-3d	PPM	0.7	23.33	1.0
AL	167.08	B	0.018	PPM	0.0007	3.88	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	6411	200
AL	167.08	B	60810	0.881
P	213.63	A	6340	197
AL	167.08	B	60808	0.881
P	213.63	A	6120	191
AL	167.08	B	59760	0.865

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
B	GH	3	07:37	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	196	PPM	4.5	2.29	1.0
AL	167.08	B	0.875	PPM	0.0092	1.05	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	6207	193
AL	167.08	B	62246	0.903
P	213.63	A	6198	193
AL	167.08	B	63122	0.916
P	213.63	A	6287	196
AL	167.08	B	63825	0.926

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
9	GH	3	07:39	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	194	PPM	1.7	0.87	1.0
AL	167.08	B	0.915	PPM	0.0115	1.25	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	6919	216
AL	167.08	B	12686	0.168
P	213.63	A	6989	218
AL	167.08	B	12719	0.169
P	213.63	A	7002	218
AL	167.08	B	12766	0.169

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
10	GH	3	07:41	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	217	PPM	1.2	0.55	1.0
AL	167.08	B	0.168	PPM	0.0007	0.41	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	7825	245
AL	167.08	B	17658	0.242
P	213.63	A	7669	240
AL	167.08	B	17522	0.240
P	213.63	A	7777	243
AL	167.08	B	17569	0.241

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
11	GH	3	07:42	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	242	PPM	2.5	1.03	1.0
AL	167.08	B	0.241	PPM	0.0010	0.41	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	6271	195
AL	167.08	B	19292	0.266
P	213.63	A	6287	196
AL	167.08	B	19562	0.270
P	213.63	A	6378	199
AL	167.08	B	19759	0.273

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
12	GH	3	07:44	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	196	PPM	2.1	1.07	1.0
Al	167.08	B	0.269	PPM	0.0035	1.30	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	69	-1
AL	167.08	B	807	-0.006
P	213.63	A	55	-2
AL	167.08	B	783	-0.006
P	213.63	A	54	-2
AL	167.08	B	787	-0.006

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
13	GH	3	07:45	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	-2d	PPM	0.7	35.00	1.0
AL	167.08	B	-0.006	PPM	0.0000	0.00	3.0

EL	NM	C	INTENSITY	CONC
P	213.63	A	39	-2
AL	167.08	B	28089	0.395
P	213.63	A	33	-2
AL	167.08	B	28066	0.395
P	213.63	A	38	-2
AL	167.08	B	27833	0.392

P 300 124660-00 14 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
40	GH	3	07:47	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
P	213.63	A	-2d	PPM	0.0	0.00	1.0
AL	167.08	B	0.394	PPM	0.0017	0.43	3.0

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

Page Ref. 0605

BUTTERWORTH LABORATORIES LIMITED
ANALYTICAL DATA RECORD SHEET

Name of Client: Huntingdon Life Sciences

BLL Reference(s): 11/0366(95) to 11/0373(95 (composite teeth
(check Customer Ref. etc.)

Methodology: Fusion in Na_2CO_3/H_3BO_3 → 25 ml in HCl

Instrumentation: ICP
(Calibration Status etc.)

Quality Control Employed: Control standard, Duplicate analysis

Reference Materials: 1000 ppm Al standard ex BDH
(If required)

Analytical Data:

Dish No	Sample wt/mg	mg/l Al	% Al
1	18.2	0.076	0.010
2	24.1	0.099	0.010
3	Blank	0.009 (<0.01)	

Analyst..... Date 27/2/96

Checked by..... Date 28/2/96

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11[illegible] TEETH

P 300 124660-00 27 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
RECAL	GH	3	08:02	AL-LOW

EL	NM	CH	SEC
AL	167.08	B	3.0

B#	INTENSITY	RAW-CONC	CONC
99	58	0.000d	0.000
1	71204	1.000	0.999

0 -62 x 61 x2

EL	NM	C	INTENSITY	CONC
AL	167.08	B	76695	1.080
AL	167.08	B	77595	1.093
AL	167.08	B	78746	1.111

P 300 124660-00 27 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
10	GH	3	08:04	AL-LOW

1 ppm in H_2O

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	1.094	PPM	0.0155	1.41	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	72715	1.021
AL	167.08	B	72485	1.017
AL	167.08	B	69536	0.973

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/1/00 (V3)

P 300 124660-00 27 FEB 96

Matrix matched

SAMPLE	OPERATOR	PWR	TIME	NAME
1	GH	3	08:05	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	1.003	PPM	0.0266	2.65	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	5915	0.077
AL	167.08	B	5911	0.077
AL	167.08	B	5867	0.076

P 300 124660-00 27 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
1	GH	3	08:07	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	0.076	PPM	0.0007	0.92	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	7627	0.100
AL	167.08	B	7523	0.098
AL	167.08	B	7697	0.101

P 300 124660-00 27 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
2	GH	3	08:08	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	0.099	PPM	0.0015	1.51	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	860	0.010
AL	167.08	B	808	0.009
AL	167.08	B	851	0.010

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

P 300 124660-00 27 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
3	GH	3	08:11	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	0.009	PPM	0.0007	7.77	3.0

EL	NM	C	INTENSITY	CONC
AL	167.08	B	71834	1.006
AL	167.08	B	71887	1.007
AL	167.08	B	71509	1.002

P 300 124660-00 27 FEB 96

SAMPLE	OPERATOR	PWR	TIME	NAME
1	GH	3	08:13	AL-LOW

EL	NM	CH	CONC	UNIT	SD	RSD	SEC
AL	167.08	B	1.005	PPM	0.0026	0.25	3.0

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

Page Ref. 0606

BUTTERWORTH LABORATORIES LIMITED
ANALYTICAL DATA RECORD SHEET

Name of Client: Huntingdon Life Sciences

BLL Reference(s): 11/0366 - 11/0373(95) Composite bone
(check Customer Ref. etc.)

Methodology: Fluoride

Instrumentation: ISE see P. 0599
(Calibration Status etc.)

Quality Control Employed: Replicate analysis

Reference Materials: Analar Sodium Fluoride
(If required)

Analytical Data:

Samples made to a final volume of 25 ml

Sample No	Wt in mg	meter reading/mV	mg/L	mg/kg
1	Blank	-30	<0.01	
2	32.74	-92	0.56	430
3	41.26	-98	0.72	440
4	27.91	-89	0.49	410
5	37.17	-92	0.56	380
6	23.92	-84	0.39	410
7	19.18	-83	0.38	500 (outlier result. Not included in calculation)
8	21.51 + 0.9ppm spike	-112	1.31	difference = 0.34 mg/L
9	— 0.9ppm spike	-105	0.97	= and 400 mg/kg

Calibration data

0.01 mg/L F	-37
0.11 mg/L F	-56
0.43 mg/L F	-86
4.33 mg/L F	-142

Average result 0.041 %

Spike recovery 98%

Analyst..... Date 13-2-96

Checked by..... Date 15-2-96

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

Page Ref. 0607

BUTTERWORTH LABORATORIES LIMITED
ANALYTICAL DATA RECORD SHEET

Name of Client: Huntingdon Life Sciences

BLL Reference(s): 11/0366 to 11/0373 Composite teeth
(check Customer Ref. etc.)

Methodology: Fluoride

Instrumentation: ISE see P 0597
(Calibration Status etc.)

Quality Control Employed: Replicate analysis

Reference Materials: Analar grade Sodium Fluoride
(If required)

Analytical Data:

Samples made to a final volume of 25 mls

2	weight/mg	reading/mV	mg/L	mg/kg
1	21.3	-62	0.17	200
2	25.5	-67	0.22	220
3	16.0	-57	0.14	220

.0 mg/L	F	-143
.5 mg/L	F	-86
.1 mg/L	F	-50

subsample of flask one was diluted with a standard to give a spike solution equivalent to 0.2 mg/L

Reading - 78 = 0.35 mg/L

Difference = 0.18 90 % recovery

Analyst..... Date 28/2/96

Checked by..... Date 5/3/96

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

QUALITY ASSURANCE REPORT

The QA unit has audited all analytical results and undertaken periodic monitoring of the Study. The dates of these audits are listed:

Date	Aspect Monitored
21 December 1995	Protocol
22 January 1996	Analytical Results
17 May 1996	Draft Report
31 May 1996	Final Report

SUMMARY

All aspects of this Study have been carried out in accordance with the Protocol. Authorised copies of data recorded in laboratory notebooks have been used which is a minor deviation from the protocol, but does not affect the validity of the analytical results.

QUALITY ASSURANCE STATEMENT

All analyses for this Study were carried out in compliance with the Principles of Good Laboratory Practice (GLP)as set out forth in 'Good Laboratory Practice, the United Kingdom Compliance Programme, Department of Health and Social Security, 1986, and subsequent revision, Department of Health, 1989'.

Analytical work carried out under this Study has also been carried out in accordance with our NAMAS accreditation.

Signed by J. Welch ..... Date 31/5/96 .....

JOHN A S WELCH CChem MRSC
Registered Analytical Chemist
Laboratory and Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 22

(Validation of method for the analysis of fluoride and aluminium contact of bone of teeth - continued)

Study Schedule No BL 11/0366 (95)

QUALITY POLICY STATEMENT

The Quality Policy of Butterworth Laboratories Limited is to provide a highly confidential and comprehensive service of analytical chemistry and to offer consultancy and expert advice in the relevant areas of pure and applied chemistry which meet today's stringent requirements for international scientific business activities.

The system developed to comply with this policy is described in the Quality Manual. It is the responsibility of all staff to be familiar with the contents of this Quality Manual and to comply with the policies and procedures described therein and with associated documentation at all times.

I the undersigned accept the responsibility that in this GLP Study this Quality Policy has been implemented.

Signed .....
DORIS E BUTTERWORTH C Chem FRSC FInstPet FInstD
Managing and Technical Director
for Butterworth Laboratories Limited

Date 31st May 1996

APPENDIX 23

Determination of aluminium and fluoride in rat teeth, bone and urine

SPONSOR

HUNTINGDON LIFE SCIENCES
(HLS)

STUDY SCHEDULE

BGH/57

STUDY OBJECTIVE

Determination of Aluminium and Fluoride
in Rat Teeth, Bone and Urine

TESTING FACILITY

BUTTERWORTH LABORATORIES LIMITED
(BLL)

STUDY SCHEDULE NUMBER

BL 6/0805 (96)

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL 6/0805 (96)

STATEMENT OF DATA CONFIDENTIALITY

Authorised copies of all data generated by Butterworth Laboratories Limited will be held in our archives for a period of not less than SIX years. The original data being transferred within the final report to the Sponsor.

As with all work undertaken by Butterworth Laboratories Limited, for Huntingdon Life Sciences Limited, customer confidentiality is observed in accordance with our Confidentiality Agreement, signed by us on 4th January 1993.

STUDY DIRECTOR'S AUTHENTICATION

I, accept responsibility for the conduct of this study and confirm that analyses were undertaken in compliance with the Principles of Good Laboratory Practice.

Signed by.....
(Study Director)

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL 6/0805 (96)

CONTENTS

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0805 (96)

PERSONNEL INVOLVED

DAVID A RICHES BSc, C.Chem, MRSC
Study Director

DAVID J HAWKINS BSc, C.Biol, MIBiol
Chemist in Charge

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL 6/0805 (96)

INTRODUCTION

Samples of terminal urine, bone and teeth were submitted for analysis on 12 November 1996. Withdrawal samples were later submitted on 13 February 1997.

All samples were analysed for Fluoride using ion selective electrode and Aluminium using inductively coupled plasma atomic emission spectrometry (ICPAES).

EXPERIMENTAL PROCEDURE

Equipment/instrumentation

Allied Analytical Instruments Plasma 300 ICP
Orion 920A meter
Fluoride electrode BLE 1
Double junction reference electrode BLE 6
Silica basin
Muffle furnace at 500°C
Small Pestle and Mortar
Water bath
Magnetic Stirrer with stirring fleas
Plastic beakers 25-40 ml
Analytical Balance (5 figure)
Grade A volumetric glassware
Plastic volumetric flasks
Platinum thimbles

Reagents and Standards

1009 mg/L Fluoride standard solution (BL ref IC041) supplied by Fischer Scientific product J/4548/05

1000 mg/L Aluminium standard solution (BL ref SP001) supplied by Merck product 140314S

Total Ionic Strength Adjustment Buffer (TISAB) supplied by Merck product 160847G

AnalaR anhydrous Sodium carbonate supplied by Merck product 102405Y

Boric acid supplied by Aldrich product No. 33,906-7

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No. BL 6/0805 (96)

Reagents and Standards (continued)

1+1 Hydrochloric acid (prepared from AnalaR grade hydrochloric acid. Merck product 102405Y

0.25N Hydrochloric acid (prepared from 1N Hydrochloric acid Merck product 190686W)

0.125N Sodium hydroxide (prepared from 1N Sodium hydroxide solution Merck product 191396T)

Analytical Procedure

Urine Samples

The urine samples were analysed for Fluoride in accordance with Butterworth Laboratories Limited documented in-house method BLM 147.

The samples were diluted 2mls to 10mls in TISAB. The Fluoride ion was detected using a Fluoride ion selective electrode.

The Aluminium was determined by ICPAES on samples which were pretreated by digestion with Sulphuric and Nitric acid in accordance with Butterworth Laboratories Limited documented in-house method BLM 58.

2ml of sample was digested and made up to 10mls with deionised water.

Copies of all the in-house methods used are contained in the appendix of this report. The results can be found on the Certificates of Analysis in this report.

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No. BL 6/0805 (96)

Bone and Teeth Samples

The bone and teeth samples were analysed in accordance with Butterworth Laboratories Limited documented in-house procedure BLM 225.

The bones and teeth were transferred to silica basins and ashed overnight in a furnace at 500°C. The ash was ground in a pestle and mortar and transferred to plastic sample pots.

Fluoride was determined by dissolution of the ground material in Hydrochloric acid. Fluoride ions were detected using an ion selective electrode.

Aluminium was determined by fusing the ground material in a mixture of Sodium carbonate and Boric acid. After acidification the solution was analysed for Aluminium by ICPAES.

The results are expressed as percent by weight of the determinand in the ashed and ground sample.

RESULTS/DISCUSSION

Urine

Fluoride measurements were being made towards the lower end of the calibration range. Duplicate determinations were within ± 10% of the mean and analyses of spiked samples gave recoveries between 98 and 102%.

There was sediment present in all the samples. The results relate to Fluoride ions in solution only.

Aluminium measurements were made on a pretreated sample. The samples were digested in Sulphuric and Nitric acid to produce a clear solution. The results therefore relate to total Aluminium in the sample.

Some difficulties in the analysis were caused by the high concentration of Phosphorus in the samples. A slight overlap of an adjacent Phosphorus emission peak may cause a slight positive bias to the results. Tests with Aluminium standards spiked with Phosphorus showed that 250ppm Phosphorus would raise the Aluminium signal by 0.02ppm (equivalent to 0.1ppm in the urine samples). A 500ppm Phosphorus spike increased the signal by 0.05ppm (equivalent to 0.25ppm in the urine samples). The presence of this interference prevented detection for Aluminium at low concentrations. A limit of quantitation of 0.5ppm was quoted for most samples, however, in a few cases, it was not possible to detect Aluminium at higher concentrations than this and so higher limits of quantitation have been quoted on the Certificate of Analysis.

Samples spiked with Aluminium gave recoveries in the range 96 to 100%. Duplicate preparations of samples gave results within ± 10% of the mean.

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No. BL 6/0805 (96)

Bone and Teeth

Fluoride measurements were again being made towards the lower end of the calibration range. Duplicate determinations were generally with ± 10% of content. The first set of data (terminal samples) were analysed using 25mg subsamples. When the withdrawal samples were analysed, 100mg subsamples were tested with a corresponding increase in the volumes of reagents to allow a more representative sample to be taken. An improvement in the agreement of duplicates was noticed with these samples.

The Aluminium determination suffered the same interference effects due to Phosphorus as observed in the urine samples. However, no Aluminium was detected in the samples above the limit of quantitation.

CONCLUSION

Low concentrations of Fluoride and Aluminium have been detected in the samples and a pattern of results has become apparent amongst some of the animal groups.

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

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Telephone: 0181-977 0750
Fax: 0181-943 2624

Study Schedule No BL6/0805 (96)

Certificate of Analysis

Our ref: BL 11/0788 (96)
JASW/jhs/DAR

4 April 1997

Huntingdon Life Sciences
P O Box 2
Huntingdon
Cambridgeshire
PE18 6ES

For the Attention of Mr D W Coombs

Samples of:	Rat Urine
BLL Study Schedule No:	BL 6/0805 (96)
A subsection of HLS Study:	BGH/57
Samples Received:	12 November 1996
Samples Analysed:	22 - 27 January 1997

BLL Reference	HLS Reference			Fluoride expressed as F	Aluminium expressed as Al
				Results in mg/Litre	
BL 11/0788 (96)	Cage 1	Gp1 1-5	5.11.96	1.4	<0.5
BL 11/0789 (96)	Cage 2	Gp1 6-10	5.11.96	1.6	2.8
BL 11/0790 (96)	Cage17	Gp1 81-85	5.11.96	1.5	<0.5
BL 11/0791 (96)	Cage18	Gp1 86-90	5.11.96	1.2	<0.7
BL 11/0792 (96)	Cage 5	Gp2 21-25	5.11.96	6.0	<0.5, <0.5
BL 11/0793 (96)	Cage 6	Gp2 26-30	5.11.96	6.5	0.8
BL 11/0794 (96)	Cage 21	Gp2 101-105	5.11.96	6.0	<0.5
BL 11/0795 (96)	Cage 22	Gp2 106-110	5.11.96	8.5	<0.6

NAMAS TESTING No 0215

GLP Compliant Laboratory

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0805 (96)

BL 11/0788 (96) continued

BLL Reference	HLS Reference				Fluoride expressed as F	Aluminium expressed as Al
					Results in mg/Litre	
BL 11/0796 (96)	Cage 9	Gp3	41-45	5.11.96	1.4 1.2	<0.5
BL 11/0797 (96)	Cage 10	Gp3	46-50	5.11.96	1.1	<0.5
BL 11/0798 (96)	Cage 25	Gp3	121-125	5.11.96	1.4	1.8
BL 11/0799 (96)	Cage 26	Gp3	126-130	5.11.96	1.4	3.2
BL 11/0800 (96)	Cage 11	Gp4	51-55	5.11.96	1.1 1.1	<0.5
BL 11/0801 (96)	Cage 12	Gp4	56-60	5.11.96	1.7	0.9
BL 11/0802 (96)	Cage 27	Gp4	131-135	5.11.96	1.4	0.7
BL 11/0803 (96)	Cage 28	Gp4	136-140	5.11.96	1.6	1.5
BL 11/0804 (96)	Cage 13	Gp5	61-65	5.11.96	3.4 3.2	<0.5
BL 11/0805 (96)	Cage 14	Gp5	66-70	5.11.96	4.2	3.4 3.0
BL 11/0806 (96)	Cage 29	Gp5	141-145	5.11.96	5.5	<0.7
BL 11/0807 (96)	Cage 30	Gp5	146-150	5.11.96	3.7	4.9 4.6

Samples analysed in accordance with Butterworth Laboratories Limited documented in-house methods BLM 58 and BLM 147.

DAVID A RICHES
Senior Manager

JOHN A S WELCH
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Butterworth Laboratories Ltd
Analytical & Consulting Chemists

54-56 Waldegrave Road,
Teddington, Middlesex
TW11 8LG UK

Telephone: 0181-977 0750
Fax: 0181-943 2624

Study Schedule No BL6/0805 (96)

Certificate of Analysis

Our ref: BL 11/0808 (96)
JASW/jhs/DAR

4 April 1997

Huntingdon Life Sciences
P O Box 2
Huntingdon
Cambridgeshire
PE18 6ES

For the Attention of Mr D W Coombs

Samples of:	Rat Femur
BLL Study Schedule No:	BL 6/0805 (96)
A subsection of HLS Study:	BGH/57
Samples Received:	12 November 1996
Samples Analysed:	13 February - 6 March 1997

BLL Reference	HLS Reference			Fluoride expressed as F Results in % m/m	Aluminium expressed as Al
BL 11/0808 (96)	Gp 1	1-5	7.11.96	0.029	<0.01
BL 11/0809 (96)	Gp1	6-10	7.11.96	0.023	<0.01
BL 11/0810 (96)	Gp1	81-85	7.11.96	0.031	<0.01
BL 11/0811 (96)	Gp1	86-90	7.11.96	0.033	<0.01
BL 11/0812 (96)	Gp2	21-25	7.11.96	0.053	<0.01
BL 11/0813 (96)	Gp2	26-30	7.11.96	0.060	<0.01
BL 11/0814 (96)	Gp2	101-105	7.11.96	0.082 0.086	<0.01
BL 11/0815 (96)	Gp2	106-110	7.11.96	0.076 0.090	<0.01

NAMAS TESTING No 0215

GLP Compliant Laboratory

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0025 (96)

BL 11/0808 (96) continued

BLL Reference	HLS Reference			Fluoride expressed as F Results in % m/m	Aluminium expressed as Al
BL 11/0816 (96)	Gp3	41-45	7.11.96	0.023	<0.01
BL 11/0817 (96)	Gp3	46-50	7.11.96	0.021	<0.01
BL 11/0818 (96)	Gp3	121-125	7.11.96	0.027	<0.01 <0.01
BL 11/0819 (96)	Gp3	126-130	7.11.96	0.032	<0.01
BL 11/0820 (96)	Gp4	51-55	7.11.96	0.020	<0.01
BL 11/0821 (96)	Gp4	56-60	7.11.96	0.015 0.018	<0.01
BL 11/0822 (96)	Gp4	131-135	7.11.96	0.025	<0.01
BL 11/0823 (96)	Gp4	136-140	7.11.96	0.031	<0.01
BL 11/0824 (96)	Gp5	61-65	7.11.96	0.040	<0.01
BL 11/0825 (96)	Gp5	66-70	7.11.96	0.042	<0.01
BL 11/0826 (96)	Gp5	141-145	7.11.96	0.063	<0.01
BL 11/0827 (96)	Gp5	146-150	7.11.96	0.063	<0.01

Samples analysed in accordance with Butterworth Laboratories Limited documented in-house methods BLM 225.

DAVID A RICHES
Senior Manager

JOHN A S WELCH
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Butterworth Laboratories Ltd
Analytical & Consulting Chemists

54-56 Waldegrave Road,
Teddington, Middlesex
TW11 8LG UK

Telephone: 0181-977 0750
Fax: 0181-943 2624

Study Schedule No BL6/0805 (96)

Certificate of Analysis

Our ref: BL 11/0828 (96)
JASW/jhs/DAR

4 April 1997

Huntingdon Life Sciences
P O Box 2
Huntingdon
Cambridgeshire PE18 6ES

For the Attention of Mr D W Coombs

Samples of:	Rat Teeth (lower jaw)
BLL Study Schedule No:	BL 6/0805 (96)
A subsection of HLS Study:	BGH/57
Samples Received:	12 November 1996
Samples Analysed:	13 February - 6 March 1997

BLL Reference	HLS Reference			Fluoride expressed as F	Aluminium expressed as Al
				Results in % m/m	
BL 11/0828 (96)	Gp 1	1-5	7.11.96	0.019	<0.01
BL 11/0829 (96)	Gp1	6-10	7.11.96	0.011	<0.01
BL 11/0830 (96)	Gp1	81-85	7.11.96	0.016	<0.01 <0.01
BL 11/0831 (96)	Gp1	86-90	7.11.96	0.020	<0.01
BL 11/0832 (96)	Gp2	21-25	7.11.96	0.019	<0.01
BL 11/0833 (96)	Gp2	26-30	7.11.96	0.020 0.022	<0.01
BL 11/0834 (96)	Gp2	101-105	7.11.96	0.025	<0.01
BL 11/0835 (96)	Gp2	106-110	7.11.96	0.010	<0.01

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0005 (96)

BL 11/0828 (96) continued

BLL Reference	HLS Reference			Fluoride expressed as F Results in % m/m	Aluminium expressed as Al
BL 11/0836 (96)	Gp3	41-45	7.11.96	0.010	<0.01
BL 11/0837 (96)	Gp3	46-50	7.11.96	0.008	<0.01
BL 11/0838 (96)	Gp3	121-125	7.11.96	0.012	<0.01
BL 11/0839 (96)	Gp3	126-130	7.11.96	0.013	<0.01
BL 11/0840 (96)	Gp4	51-55	7.11.96	0.017 0.018	<0.01
BL 11/0841 (96)	Gp4	56-60	7.11.96	0.016	<0.01
BL 11/0842 (96)	Gp4	131-135	7.11.96	0.018	<0.01
BL 11/0843 (96)	Gp4	136-140	7.11.96	0.018	<0.01
BL 11/0844 (96)	Gp5	61-65	7.11.96	0.017	<0.01
BL 11/0845 (96)	Gp5	66-70	7.11.96	0.017	<0.01
BL 11/0846 (96)	Gp5	141-145	7.11.96	0.022	<0.01
BL 11/0847 (96)	Gp5	146-150	7.11.96	0.024	<0.01 <0.01

Samples analysed in accordance with Butterworth Laboratories Limited documented in-house methods BLM 225.

DAVID A RICHES
Senior Manager

JOHN A S WELCH
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Butterworth Laboratories Ltd
Analytical & Consulting Chemists

54-56 Waldegrave Road,
Teddington, Middlesex
TW11 8LG UK

Telephone: 0181-977 0750
Fax: 0181-943 2624

Study Schedule No BL6/0805 (96)

Certificate of Analysis

Our Ref: BL 2/0783 (96)
JASW/jhs/DAR

23 April 1997

Huntingdon Life Sciences
P O Box 2
Huntingdon
Cambridgeshire
PE18 6ES

For the Attention of Mr D W Coombs

Samples of:	Rat Femur
BLL Study Schedule No:	BL 6/0805 (96)
A subsection of HLS Study:	BGH/57
Samples Received:	13 February 1997
Samples Analysed:	17 March - 11 April 1997

BLL Reference	HLS Reference		Fluoride expressed as F	Aluminium expressed as Al
			Results in % m/m	
BL 2/0783 (97)	Group 1	11-15	0.027	<0.01
BL 2/0786 (97)	Group 1	16-20	0.026	<0.01
				<0.01
BL 2/0789 (97)	Group 1	91-95	0.036	<0.01
BL 2/0792 (97)	Group 1	96-100	0.038	<0.01
BL 2/0795 (97)	Group 2	31-35	0.058	<0.01
BL 2/0798 (97)	Group 2	36-40	0.050	<0.01
BL 2/0801 (97)	Group 2	111-115	0.075	<0.01
BL 2/0804 (97)	Group 2	116-120	0.070	<0.01

NAMAS TESTING No 0215

GLP Compliant Laboratory

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0805 (96)

BL 2/0783 (97) continued

BLL Reference	HLS Reference		Fluoride expressed as F	Aluminium expressed as Al
			Results in % m/m	
BL 2/0807 (97)	Group 5	71-75	0.038 0.038	<0.01
BL 2/0810 (97)	Group 5	76-80	0.040	<0.01 <0.01
BL 2/0813 (97)	Group 5	151	0.052	<0.01
BL 2/0815 (97)	Group 5	152-155	0.064 0.064	<0.01
BL 2/0818 (97)	Group 5	156-160	0.051	<0.01

Samples analysed in accordance with Butterworth Laboratories Limited documented in-house methods BLM 225.

DAVID A RICHES
Senior Manager

JOHN A S WELCH
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Butterworth Laboratories Ltd
Analytical & Consulting Chemists

54-56 Waldegrave Road,
Teddington, Middlesex
TW11 8LG UK

Telephone: 0181-977 0750
Fax: 0181-943 2624

Study Schedule No BL6/0805 (96)

Certificate of Analysis

Our Ref: BL 2/0784 (97)
JASW/jhs/DAR

23 April 1997

Huntingdon Life Sciences
P O Box 2
Huntingdon
Cambridgeshire
PE18 6ES

For the Attention of Mr D W Coombs

Samples of:	Rat Urine
BLL Study Schedule No:	BL 6/0805 (96)
A subsection of HLS Study:	BGH/57
Samples Received:	13 February 1997
Samples Analysed:	19 - 26 March 1997

BLL Reference	HLS Reference		Fluoride expressed as F	Aluminium expressed as Al
			Results in mg/L	
BL 2/0784 (97)	Group 1	11-15	1.0	<0.5 <0.5 0.6
BL 2/0787 (97)	Group 1	16-20	1.1	<0.5
BL 2/0790 (97)	Group 1	91-95	1.2	<0.5
BL 2/0793 (97)	Group 1	96-100	0.96 0.91	<0.5
BL 2/0796 (97)	Group 2	31-35	1.6	<0.5
BL 2/0799 (97)	Group 2	36-40	1.4	<0.5 <0.5
BL 2/0802 (97)	Group 2	111-115	1.3	0.8

NAMAS TESTING No. 0215

GLP Compliant Laboratory

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0005 (96)

BL 2/0784 (97) continued

BLL Reference	HLS Reference		Fluoride expressed as F	Aluminium expressed as Al
			Results in mg/L	
BL 2/0805 (97)	Group 2	116-120	1.3	<0.6
BL 2/0808 (97)	Group 5	71-75	0.9	<0.5
BL 2/0811 (97)	Group 5	76-80	1.0 1.0	<0.5
BL 2/0816 (97)	Group 5	152-155	1.2	<0.5
BL 2/0819 (97)	Group 5	156-160	1.2	<0.5

Samples analysed in accordance with Butterworth Laboratories Limited documented in-house methods BLM 147.

DAVID A RICHES
Senior Manager

JOHN A S WELCH
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Butterworth Laboratories Ltd
Analytical & Consulting Chemists

54-56 Waldegrave Road,
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Telephone: 0181-977 0750
Fax: 0181-943 2624

Study Schedule No BL6/0805 (96)

Certificate of Analysis

Our Ref: BL 2/0782 (97)
JASW/jhs/DAR

23 April 1997

Huntingdon Life Sciences
P O Box 2
Huntingdon
Cambridgeshire
PE18 6ES

For the Attention of Mr D W Coombs

Samples of:	Rat Teeth
BLL Study Schedule No:	BL 6/0805 (96)
A subsection of HLS Study:	BGH/57
Samples Received:	13 February 1997
Samples Analysed:	17 March - 11 April 1997

BLL Reference	HLS Reference		Fluoride expressed as F	Aluminium expressed as Al
			Results in % m/m	
BL 2/0782 (97)	Group 1	11-15	0.014	<0.01
BL 2/0785 (97)	Group 1	16-20	0.012	<0.01
BL 2/0788 (97)	Group 1	91-95	0.015	<0.01
BL 2/0791 (97)	Group 1	96-100	0.016 0.017	<0.01
BL 2/0794 (97)	Group 2	31-35	0.011	<0.01
BL 2/0797 (97)	Group 2	36-40	0.010	<0.01 <0.01
BL 2/0800 (97)	Group 2	111-115	0.012	<0.01
BL 2/0803 (97)	Group 2	116-120	0.014	<0.01

NAMAS TESTING No. 0215

GLP Compliant Laboratory

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL6/0005 (96)

BL 2/0782 (97) continued

BLL Reference	HLS Reference		Fluoride expressed as F	Aluminium expressed as Al
			Results in % m/m	
BL 2/0806 (97)	Group 5	71-75	0.009	<0.01
BL 2/0809 (97)	Group 5	76-80	0.010	<0.01
BL 2/0812 (97)	Group 5	151	0.028	<0.01
BL 2/0814 (97)	Group 5	152-155	0.014	<0.01
BL 2/0817 (97)	Group 5	156-160	0.011	<0.01

Samples analysed in accordance with Butterworth Laboratories Limited documented in-house methods BLM 225.

DAVID A RICHES
Senior Manager

JOHN A S WELCH
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

PROTOCOL

SCHEDULE NO:

BL 6/0805 (96)

SPONSOR

Huntingdon Life Sciences Limited
PO Box 2, Huntingdon, Cambridgeshire PE18 6ES

TEST MATERIAL

32 samples of Rat Bone, Teeth and Urine Samples

OBJECTIVE OF STUDY

Determination of Aluminium and Fluoride in the test materials.

TESTING FACILITY

Butterworth Laboratories Limited
54-56 Waldegrave Road, Teddington, Middlesex TW11 8LG

STARTING DATE

July 1996

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

COMPLETION DATE

Analysis to be completed within 15 working days from receipt of samples.

A copy of the Certificate of Analysis will be issued as an interim report, with the original Certificate of Analysis being bound in the final GLP report to be issued within 25 working days of completion of the analytical work.

METHODS TO BE USED

Butterworth Laboratories Limited documented In-house methods BLM 147 "The determination of Fluoride by Ion Selective Electrode in Rat Urine" and BLM 225 "The determination of Aluminium and Fluoride in Tooth and Bone samples.

Aluminium in urine will be analysed directly by ICP after dilution in accordance with SOP IM66.

RECORDS TO BE ARCHIVED

All study specific raw data Instrumentation used, calibration standard results, all readings generated, all relevant sample/batch and reference numbers. Authorised copies of relevant raw data will be included in the final Report.

STUDY DIRECTOR

David A Riches BSc CChem MRSC
Senior Manager

MONITORING SCIENTIST

Derek Coombs
Division of Toxicology
Huntingdon Life Sciences Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

ACCREDITATION

The Study will be conducted in compliance with the Principles of Good Laboratory Practice (GLP) as set forth in "Good Laboratory Practice, the United Kingdom Compliance Programme, Department of Health and Social Security, 1986, and subsequent revision, Department of Health 1989" and Good Laboratory Practice in the Testing of Chemicals OECD ISBN 92-64-12367-9, Paris 1982, subsequently republished OECD Environment Monograph No 45 1992.

Accepted on behalf of Butterworth Laboratories Limited

Signed.....
David A Riches BSc CChem MRSC
Senior Manager
Study Director

Date...25/6/96.....

Accepted on behalf of Huntingdon Life Sciences Limited

Signed.....
Derek Coombs
Division of Toxicology
Monitoring Scientist

Date 26 June 1996

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

PROTOCOL AMENDMENT

SCHEDULE NO: BL 6/0805 (96)

SPONSOR: Huntingdon Life Sciences

TESTING FACILITY: Butterworth Laboratories Limited
54-56 Waldegrave Road
Teddington
Middlesex
TW11 8LG

NATURE OF AMENDMENT: Urine samples to be digested prior to Aluminium analysis in accordance Butterworth Laboratories Limited documented in-house method BLM 58. PTFE beakers to be used instead of glass and sample volumes reduced as necessary.
Amendment required due to high concentration of suspended matter in samples.

SIGNATURES:

Signed by..... Date 17/3/97
David A Riches BSc C.Chem MRSC
Study Director

Signed by..... Date 18/3/97
Derek Coombs
Division of Toxicology
Monitoring Scientist

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL 6/0805 (96)

QUALITY ASSURANCE REPORT

The QA unit has audited all analytical results and undertaken periodic monitoring of the Study. The dates of these audits are listed:

Date	Aspect Monitored
25 June 1996	Protocol
17 March 1997	Protocol Ammendment
19 March 1997	Analytical Results
30 April 1997	Final Report

SUMMARY

All aspects of this Study have been carried out in accordance with the Protocol.

QUALITY ASSURANCE STATEMENT

All analyses for this Study were carried out in compliance with the Principles of Good Laboratory Practice (GLP)as set out forth in 'Good Laboratory Practice, the United Kingdom Compliance Programme, Department of Health and Social Security, 1986, and subsequent revision, Department of Health, 1989'.

Analytical work carried out under this study has also been carried out in accordance with our NAMAS accreditation.

Signed by J. Welch ..... Date 30/4/97 .....
JOHN A S WELCH C Chem, MRSC,
Registered Analytical Chemist
Quality Assurance Manager
for Butterworth Laboratories Limited

APPENDIX 23

(Determination of aluminium and fluoride in rat teeth, bone and urine - continued)

Study Schedule No BL 6/0805 (96)

QUALITY POLICY STATEMENT

The Quality Policy of Butterworth Laboratories Limited is to provide a highly confidential and comprehensive service of analytical chemistry and to offer consultancy and expert advice in the relevant areas of pure and applied chemistry which meet today's stringent requirements for international scientific business activities.

The system developed to comply with this policy is described in the Quality Manual. It is the responsibility of all staff to be familiar with the contents of this Quality Manual and to comply with the policies and procedures described therein and with associated documentation at all times.

I the undersigned accept the responsibility that in this GLP Study this Quality Policy has been implemented.

Signed by Doris Butterworth

DORIS E BUTTERWORTH C Chem FRSC FInstPet FInstD
Managing and Technical Director
for Butterworth Laboratories Limited

Date 30th April 1997

APPENDIX 24

Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations

GLP Final Report

IDENTITY AND ANALYTICAL MATERIAL BALANCE
REANALYSIS AFTER TOXICOLOGICAL EXAMINATIONS

Bayer AG ZF-Forschungsdienste Gebäude Q 18 51368 Leverkusen	Date: 04. November 1997 Study number: A 92/0098/02 LEV Study director: Dr. Rieckhoff Deputy study director: Dr. Neupert

Test substance: Kryolith synth. leicht gemahlen

Sponsor: Dr. Wischer, Al-Stab, LEV, O 1
Dr. Löser, PH-PDT, ELB, 431

Order no.: 92/076

Chemical name: Trinatrium-Aluminium-Hexafluorid

Empirical formula: $AlF_6 \cdot 3Na$

Molecular mass: 209.9 g/mol

CAS no.:	13775-53-6	Batch no.:	2
Sample no./year:	486448/1992	Date of production:	29 Aug 92
Producer:	AC-P3	Sampling date:	10 Sept 92
Product-no.:	002209-00		

Study initiation date: 18 Sept 1997
Study completion date: 30 Oct 1997

1 Identity

1.1 Test: Identification by FTIR
SOP.: D 0085601 DZA
Supervisor: Dr. Seelemann
Result: The FTIR spectrum is in compliance with the requirements

Procedure: Fourier Transform - Infrared Spectrometry
The $AlF_6 \cdot 3Na$ in the sample is identified by FTIR using a potassium bromide cell.
A small part of the solid sample is grinded with potassium bromide and pressed to give a transparent pellet.
The sample is analysed by high-resolution FTIR. The identification is based upon the absorption pattern and the wave numbers of the absorption maxima in the spectrum.

APPENDIX 24

(Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations - continued)

GLP Final Report

IDENTITY AND ANALYTICAL MATERIAL BALANCE
REANALYSIS AFTER TOXICOLOGICAL EXAMINATIONS

Bayer AG		Date:	04. November 1997
ZF-Forschungsdienste		Study number:	A 92/0098/02 LEV
Gebäude Q 18		Study director:	Dr. Rieckhoff
51368 Leverkusen		Deputy study director:	Dr. Neupert

Test substance: Kryolith synth. leicht gemahlen

Sponsor: Dr. Wischer, Al-Stab, LEV, O 1
Dr. Löser, PH-PDT, ELB, 431

Order no.: 92/076

2 Description of methods and results

2.1 Investigations of balancing

2.1.1 Test: Fusion decomposition
Method no.: 2011-0362801-92D
Supervisor: Dr. Rieckhoff
Result: –

Procedure: Fusion decomposition
Approximately 0.2 - 0.5 grams of the sample and 2,5 - 3 grams of the fluxing agent (litium tetraborate) were placed in a platinum crucible. This mixture is molten gently by means of a propan oxygen burner up to 1200 °C. To achieve a complete decomposition and a sufficient homogeneity the sample was held in the molten state for two minutes. For dissolution the liquid fusion is transfered directly into a PTFE beaker containing water and nitric acid.

2.1.2 Test: Sodium (ICP-OES)
Method no.: 2011-0106301-90D
Supervisor: Dr. Rieckhoff
Result: 34.3 %

Procedure: Inductively Coupled Plasma Emission Spectrometry (ICP-OES)
The sample is mineralized by fusion decomposition, dissolved in water/nitric acid and diluted with water. The sodium content is determined by the intensity measurement of the emisson signal at the sodium emission line (589.6 nm). The measured intensity is compared with the intensity of the calibration solution.

2.1.3 Test: Aluminium (ICP-OES)
Method no.: 2011-0249101-91D
Supervisor: Dr. Rieckhoff
Result: 14.4 %

Procedure: Inductively Coupled Plasma Emission Spectrometry (ICP-OES)
The sample is mineralized by fusion decomposition, dissolved in water/nitric acid and diluted with water. The aluminium content is determined by the intensity measurement of the emisson signal at the aluminium emission line (396.152 nm). The measured intensity is compared with the intensity of the calibration solution.

APPENDIX 24

(Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations - continued)

GLP Final Report

IDENTITY AND ANALYTICAL MATERIAL BALANCE
REANALYSIS AFTER TOXICOLOGICAL EXAMINATIONS

Bayer AG		Date:	04. November 1997
ZF-Forschungsdienste		Study number:	A 92/0098/02 LEV
Gebäude Q 18		Study director:	Dr. Rieckhoff
51368 Leverkusen		Deputy study director:	Dr. Neupert

Test substance: Kryolith synth. leicht gemahlen

Sponsor: Dr. Wischer, Al-Stab, LEV, O 1
Dr. Löser, PH-PDT, ELB, 431

Order no.: 92/076

2.1.4 Test: Silicon (ICP-OES)
Method no.: 2011-0299001-91D
Supervisor: Dr. Rieckhoff
Result: 0.2 %

Procedure: Inductively Coupled Plasma Emission Spectrometry (ICP-OES)
The sample is mineralized by fusion decomposition, dissolved in water/nitric acid and diluted with water. The silicon content is determined by the intensity measurement of the emisson signal at the silicon emission line (251.612 nm). The measured intensity is compared with the intensity of the calibration solution.

2.1.5 Test: Microwave assisted pressure digestion
Method no.: 2011-0440701-93D
Supervisor: Dr. Rieckhoff
Result: –

Procedure: Microwave assisted pressure digestion
This digestion is suitable for the determination of volatile elements in organic or inorganic samples. Approximately 0.1 - 0.2 grams of the sample and 4 ml of nitric and sulphuric acid were placed in a teflon vessel. Afterwards the vessels is closed, placed in the microwave oven and the power-time programm of the microwave is started. After finishing the digestion procedure the digestion solution is transfered into a volumetric flask and diluted to volume with water.

2.1.6 Test: Phosphorus (ICP-OES)
Method no.: 2011-0106001-90D
Supervisor: Dr. Rieckhoff
Result: 70 mg/kg

Procedure: Inductively Coupled Plasma Emission Spectrometry (ICP-OES)
The sample is mineralized by microwave assisted pressure digestion.. The phosphorus content is determined by the intensity measurement of the emisson signal at the phosphorus emission line (185.943 nm). The measured intensity is compared with the intensity of the calibration solution.

APPENDIX 24

(Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations - continued)

GLP Final Report

IDENTITY AND ANALYTICAL MATERIAL BALANCE
REANALYSIS AFTER TOXICOLOGICAL EXAMINATIONS

Bayer AG		Date:	04. November 1997
ZF-Forschungsdienste		Study number:	A 92/0098/02 LEV
Gebäude Q 18		Study director:	Dr. Rieckhoff
51368 Leverkusen		Deputy study director:	Dr. Neupert

Test substance: Kryolith synth. leicht gemahlen

Sponsor: Dr. Wischer, Al-Stab, LEV, O 1
Dr. Löser, PH-PDT, ELB, 431

Order no.: 92/076

2.1.7 Test: Sulphate (IC)
Method no.: 2011-0348401-92D
Supervisor: Dr. Neupert
Result: 460 mg/kg / 460 mg/kg

Procedure: Ion Chromatography
Precolumn: HPIC-AG4a
Analytical Column: HPIC-AS4a
Eluent: 0.0017 mole/L $NaHCO_3$
0.0018 mole/L Na_2CO_3
Flow: 2 ml/min
Detector: Conductivety

Quantification is made by external standardization.

2.1.8 Test: Residue on ignition
Method no.: 2011-0438303-96D
Supervisor: Dr. Königer
Result: 0.6 %

Procedure: Ignition to constant weight
The sample is placed in a porcelain crucible and heated up gently by means of an electric oven until thermal decomposition. After decomposition the residue is ignited in a muffle-furnace (800 °C) until constant weight is achieved.

2.1.9 Test: Fluoride (volumetric method)
Method no.: SOP 12.10 AC-F/367
Supervisor: Dr. Dorn, Al-F-Analytik
Result: 52.51 % / 52.43 %

Procedure: Fluoride (volumetric method)
The sample is digested using sodium carbonate. The aqueous digestion solution is placed in a steam distillation apparatus and a mixture of perchloric and phosphorus acid is added. Generated volatile fluorides (silicon tetrafluoride, hydrogen fluoride) were transfered by means of water vapor into a vessel containing a water supply. The fluoride content is determined by titration using standardized sodium hydroxide solution (indicator: bromomethylblue).

APPENDIX 24

(Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations - continued)

GLP Final Report

IDENTITY AND ANALYTICAL MATERIAL BALANCE
REANALYSIS AFTER TOXICOLOGICAL EXAMINATIONS

Bayer AG		Date:	04. November 1997
ZF-Forschungsdienste		Study number:	A 92/0098/02 LEV
Gebäude Q 18		Study director:	Dr. Rieckhoff
51368 Leverkusen		Deputy study director:	Dr. Neupert

Test substance: Kryolith synth. leicht gemahlen

Sponsor: Dr. Wischer, Al-Stab, LEV, O 1
Dr. Löser, PH-PDT, ELB, 431

Order no.: 92/076

3 Balance

measured	*theory*	*difference*
34.3 %	32.9 % sodium	+ 1.4 %
14.4 %	12.9 % aluminium	+ 1.5 %
52.5 %	54.3 % fluoride	- 1.8 %
101.2 %	100.1 %	

4 Review and comment

- The precision of used analytical methods including all sample preparation and dilution steps is about 5-10 %.
- The material balance is complete with respect to the state of the art.
- The raw data are checked and kept in archives.
- The determinations of fluoride (paragraph 2.1.9) have not been performed according to the OECD principles of Good Laboratory Practice (GLP) as described in paragraph 7.

5 Storage of records

GLP-Archiv, Bayer AG, ZF-D Zentrale Analytik Leverkusen, Gebäude O 13,
51368 Leverkusen

Study plan, raw data, final report, inspection reports and all other data relevant for verification are kept in archives.

6 Retained sample

GLP-Probenlager, Bayer AG, ZF-D Zentrale Analytik Leverkusen, Gebäude O 13,
51368 Leverkusen

APPENDIX 24

(Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations - continued)

GLP Final Report

IDENTITY AND ANALYTICAL MATERIAL BALANCE
REANALYSIS AFTER TOXICOLOGICAL EXAMINATIONS

Bayer AG ZF-Forschungsdienste Gebäude Q 18 51368 Leverkusen		Date: Study number: Study director: Deputy study director:	04. November 1997 A 92/0098/02 LEV Dr. Rieckhoff Dr. Neupert
Test substance:	Kryolith synth. leicht gemahlen		
Sponsor:	Dr. Wischer, Al-Stab, LEV, O 1 Dr. Löser, PH-PDT, ELB, 431		Order no.: 92/076

7 Declaration of study director

The investigations have been performed in accordance with the OECD principles of Good Laboratory Practice (GLP) from 04.02.83 (published in Bundesanzeiger Nr. 42 a of 02.03.83) and the principles of Good Laboratory Practice (GLP) with respect to Appendix 1 of the „Gesetz zum Schutz vor gefährlichen Stoffen" (ChemG, „Law on Protection from Hazardous Substances") from 25.07.94 (published in Bundesgesetzblatt, Teil I of 29.07.94).

Study director: 04.11.97 Rieckhoff
(Date)

Enclosure
Declaration of quality assurance

APPENDIX 24

(Re analysis certificate of sodium hexafluoroaluminate after toxicological examinations - continued)

Bayer AG
ZF-D Zentrale Analytik Leverkusen
Gebäude O 13
D-51368 Leverkusen
Federal Republic of Germany

Statement of the Quality Assurance on the Final Report

Key of the GLP-study: A 92/0098/02 LEV
Titel of the GLP-study: Kryolith synth. leicht permahlen
Identity and Analytical Material Balance
Reanalysis after Toxicological Examinations

This GLP-study was inspected by the quality assurance.
The dates of inspections and the dates of reports to the management and the study director are

inspection (date)	report (date)
18th Sep. 97	18th Sep. 97
3rd Nov. 97	3rd Nov. 97
5th Nov. 97	5th Nov. 97

The results shown in the final report on this study were inspected on the basis of the current SOPs/analytical methods. It is confirmed, that the reported results to the best of our knowledge accurately reflect the raw data of the study.

Quality assurance:

5th Nov. 97
(date)

(Dr. Dittrich/~~Dr. Häberle~~)