3M

Study Title

Oral (Gavage) Pharmacokinetic Study of PFOS in Rats

PROTOCOL

Author
Lisa Clemen

Date:
June 8, 1999

Performing Laboratory
3M Environmental Technology & Safety Services
3M Environmental Laboratory
935 Bush Avenue
St. Paul, MN 55106

Laboratory Project Identification
FACT-TOX-110
U2849

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Date 4/19/2000

Study Identification

Oral (Gavage) Pharmacokinetic Study of PFOS in Rats

Test Material	Perfluorooctane sulfonic acid potassium salt (T-6295)
Sponsor	3M Toxicology Services - Medical Department 3M Center, Building 220-2E-02 St. Paul, MN 55144-1000
Sponsor Representative	Marvin T. Case, D.V.M., Ph.D. 3M Toxicology Services Telephone: 651-733-5180 Facsimile: 651-733-1773
Study Director	Kristen J. Hansen, Ph.D. 3M Environmental Technology and Safety Services Building 2-3E-09 651-778-6018
Study Location(s)	
In vivo Testing Facility	Argus Research Laboratories, Inc. 905 Sheehy Drive, Building A Horsham, PA 19044
Analytical Testing Laboratory	3M Environmental Laboratory Building 2-3E-09 935 Bush Avenue St. Paul, MN 55106

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Sub-Contract Laboratory	Advanced Bioanalytical Services, Inc. 15 Catherwood Road Ithaca, NY 14850 Battelle Memorial Institute 505 King Avenue Columbus, Ohio 43201-2693
Proposed Study Timetable	
Study Initiation Date	June 8, 1999
Study Completion Date	August 8, 2000

1. STUDY

Oral (gavage) pharmacokinetic study of potassium perfluorooctane sulfonic acid (PFOS) in rats.

2. PURPOSE

This analytical study is designed to determine levels of potassium perfluorooctanesulfonate (PFOS) in specimens of liver and serum of rats. The in-life portion of this study was conducted at Argus Research Laboratories, study #418-013. All serum samples will be extracted and analyzed at Advanced Bioanalytical Services, Inc. and all liver samples extracted and analyzed at Battelle Memorial Institute. Additional analyses may be performed at the 3M Environmental Laboratory as methods are developed and validated. If additional analyses are performed an amendment to this protocol will be written.

3. REGULATORY COMPLIANCE

This study will be conducted in accordance with the United States Food and Drug Administration, Good Laboratory Practices Standards, Final Rule 21 CFR 58, with the exception that analysis of the test material mixture for concentration, solubility, homogeneity, and stability will not be conducted, and is the responsibility of the Sponsor.

4. QUALITY ASSURANCE

The 3M Environmental Laboratory Quality Assurance Unit will review the protocol and audit study conduct, data, and final report to determine compliance with Good Laboratory Practice Standards and with 3M Environmental Laboratory Standard Operating Procedures.

The QA Unit at the sub-contract laboratory will audit their study conduct, data, and results report prior to submitting to the 3M Environmental Laboratory.

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5. TEST MATERIAL

5.1 Refer to Argus Research Laboratory protocol for study #418-013.

6. CONTROL MATRICES

6.1 *Identification* Rat liver and serum and/or rabbit liver and serum, traceability numbers will be recorded in the raw data and included in the final report

6.2 *Source* Argus Research and/or Sigma Chemical

6.3 *Physical Description* Rat liver and serum and/or rabbit liver and serum

6.4 *Purity and Stability* Not applicable

6.5 *Storage Conditions* Frozen at -20 °C ± 10 °C or –50 °C ± 10 °C

6.6 *Reserve Matrix* A portion of the control matrix will be retained in the 3M archives for as long as the quality of the preparation affords evaluation, but not longer than ten years following the effective date of the final test rule (if applicable).

6.7 *Disposition* Matrices will be retained at the 3M Environmental Laboratory per GLP regulation. Certain matrices (feces, urine, and blood) may be disposed after QAU verification.

6.8 *Safety Precautions* Refer to MSDS for chemicals used. Wear appropriate laboratory attire, and follow adequate precautions for handling biological materials and preparing samples for analysis.

7. REFERENCE MATERIAL

7.1 *Identification* Potassium perfluorooctanesulfonate (PFOS), lot #s 171, 215, or 217 (equivalent lots)

7.2 *Source* 3M Specialty Chemicals

7.3 *Physical Description* White powder

7.4 *Purity and Stability* Purity of PFOS is 99% or greater. Stability has not been determined.

7.5 *Storage Conditions* Room temperature

7.6 *Reserve Material* A reserve sample from each batch of PFOS used in this study will be retained as long as the quality of the preparation affords evaluation, but not longer than ten years following the effective date of the final test rule (if applicable).

7.7 *Disposition* Unused reference material will be retained for use by the 3M Environmental Laboratory and will be discarded when the quality of preparation no longer affords evaluation.

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7.8 *Safety Precautions* Refer to MSDS for chemicals used. Wear appropriate laboratory attire, and follow adequate precautions for handling biological materials and preparing samples for analysis.

8. *Test System*

Rats were used as the test system and were maintained and dosed as described in the Argus Research protocol #418-013. The female rats will be given the test material or control once daily beginning 42 days prior to cohabitation and continue through day 14 or 20 of presumed gestation. See table 1 for more dosage information.

Table 1
Dosage Levels, Concentration, and Volumes

Dosage Group	Number of female rats	Dosage mg/kg/day	Concentration mg/kg/day	Dosage volume mL/kg
1	16	0	0	5
2	16	0.1	0.02	5
3	16	0.4	0.08	5
4	16	1.6	0.32	5
5	16	3.2	0.64	5

9. *Specimen and Sample Receipt*

The 3M Environmental Laboratory will receive homogeneity samples for dose analysis and specimens of the following body tissues and fluids from the indicated points in the study. See table 2 for specimen information. All specimens will be packed on dry ice for shipping.

Table 2
Specimen Information

Body tissue/fluid	Collected	Expected # of specimens
Serum – Dam and Fetus animals	Dam-Predose, Days 7, 15, and 21 Fetus-At termination of the study	320 Dam and 320 Fetus (pooled)
Urine and feces – Dam animals	Predose, Days 7, 15, and 21	320 urine and 320 feces
Liver – Dam and Fetus animals	Dam and Fetus-At termination of the study	80 Dam and 80 Fetus (pooled)
Milk Secreting Glands – Dam animals	At the termination of the study	80
Amniotic Fluid – Dam animals	Day 15 and Day 21	80

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Table 2 Cont. Specimen Information		
Body tissue/fluid	**Collected**	**Expected # of specimens**
Embryo's with Placentae – Dam animals	Day 15 and Day 21	80
Carcass – Fetus animal	At the termination of the study	160
Placentae – Fetus animal	Day 15 and Day 21	80
Liver/lung – Fetus animal	At the termination of the study	80

Total number of expected specimens: 2000
Total number of test animals: 64
Total number of control animals: 16

Specimens sent to 3M Environmental Laboratories will be received and tracked according to applicable Standard Operating Procedures.

10. PREPARATORY METHODS

10.1 FACT-M-1.1, Extraction of Potassium Perfluorooctanesulfonate or Other Anionic Fluorochemical Surfactant from Liver for Analysis Using HPLC-Electrospray/Mass Spectrometry

10.2 ETS-8-4.1, Extraction of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds from Serum or Other Fluid for Analysis Using HPLC-Electrospray/Mass Spectrometry

10.3 If preparatory methods other than those listed above are used, an amendment to this protocol will be written. Any deviations from these methods will be documented and included with the study data.

10.4 If analyses are sub-contracted to other laboratories, an amendment will be written to include their methods and copies of each method will be attached to this protocol.

11. ANALYTICAL METHODS

11.1 FACT-M-2.1, Analysis of Fluorochemicals in Liver Extracts Using HPLC-Electrospray/Mass Spectrometry

11.2 ETS-8-5.1, Analysis of Potassium Perfluorooctanesulfonate or Other Fluorochemicals in Serum or Other Fluid Extracts Using HPLC-Electrospray/Mass Spectrometry

11.3 If analytical methods other than those listed above are used, an amendment to this protocol will be written. Any deviations from these methods will be documented and included with the study data.

11.4 If analyses are sub-contracted to other laboratories, an amendment will be written to include their methods and copies of each method will be attached to this protocol.

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12. DATA QUALITY OBJECTIVES

The number of spikes/duplicates, use of surrogates, and information on other data quality indicators are included in the analytical methods. In addition, the following criteria will be met:

12.1 Linearity $r^2 \geq 0.98$

12.2 Limits of detection / quantitation

12.2.1 Method Detection Limit (MDL) for PFOS

a) Serum: 1.75 ppb

b) Liver: 15 ppb

12.2.2 Limit of Quantitation (LOQ) – Equal to the lowest acceptable standard in the calibration curve

12.3 Duplicate acceptable precision < 30% for the method

12.4 Spike acceptable recoveries 70% - 130%

12.5 Use of confirmatory methods Indeterminate samples will be re-analyzed using a confirmatory method. If a confirmatory method is used, an amendment to this protocol will be written.

12.6 Demonstration of specificity Chromatographic retention time, mass spectral daughter ion characterization.

13. SUB-CONTRACTED ANALYSIS

13.1 All analyses as detailed in this protocol will be performed at 3M Environmental Laboratories, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55106, at Advanced Bioanalytical Services, Inc., 15 Catherwood Road, Ithaca, NY 14850, or at Battelle Memorial Institute, 505 King Avenue, Columbus, Ohio 43201-2693.

13.2 An amendment to this protocol will be written if analyses are performed at laboratories other than 3M Environmental Laboratories, Advanced Bioanalytical Services, Inc., or Battelle Memorial Institute.

14. STATISTICAL ANALYSIS

Averages and standard deviations will be calculated. The statistical methods that will be used are described below:

14.1 Data transformations and analysis Data will be reported as the concentration (weight/weight or weight/vol) of PFOS or metabolite per tissue or fluid.

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14.2 *Statistical analysis* Statistics used may include regression analysis of concentrations over time, and standard deviations calculated for the concentrations within each dose group. If necessary, simple statistical tests, such as Student's t test, may be applied to evaluate statistical difference.

15. REPORT

A report containing all the results of the study will be prepared by the 3M Environmental Laboratory. If analyses are sub-contracted to other laboratories, each laboratory will prepare a report and submit it to the 3M Environmental Laboratory for inclusion in the 3M Environmental Laboratory report. Each report will include, but not be limited to, the following, when applicable:

15.1 Name and address of the facility performing the study

15.2 Dates upon which the study was initiated and completed

15.3 A statement of compliance by the Study Director addressing any exceptions to Good Laboratory Practice Standards

15.4 Objectives and procedures as stated in the approved protocol, including any changes in the original protocol

15.5 The test substance identification by name, chemical abstracts number or code number, strength, purity, and composition or other appropriate characteristics, if provided by the Sponsor

15.6 Stability and the solubility of the test substances under the conditions of administration, if provided by the Sponsor

15.7 A description of the methods used to conduct the test(s)

15.8 A description of the test system

15.9 A description of any circumstances that may have affected the quality or the integrity of the data

15.10 The name of the Study Director and the names of other scientists, professionals, and supervisory personnel involved in the study

15.11 A description of the transformations, calculations, or operations performed on the data, a summary and analysis of the analytical chemistry data, and a statement of the conclusions drawn from the analyses

15.12 Statistical methods used to evaluate the data, if applicable

15.13 The signed and dated reports of each of the individual scientists or other professionals involved in the study, if applicable

15.14 The location where raw data and the final report are to be stored

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15.15 A statement prepared by the Quality Assurance Unit listing the dates that study inspections and audits were made, and the dates of any findings reported to the Study Director and Management

If it is necessary to make corrections or additions to a report after it has been accepted, the changes will be made in the form of an amendment issued by the Study Director. The amendment will clearly identify the part of the report that is being amended, the reasons for the amendment, and will be signed by the Study Director.

16. Location of Raw Data, Records, and Final Report

Original data, or copies thereof, will be available at the 3M Environmental Laboratory to facilitate audits of the study during its progress and before acceptance of the final report. When the final report is completed, all original paper data, including those items listed below, will be retained in the archives of 3M Environmental Laboratory for at least a period of time as specified by regulation, and as established by 3M Environmental Laboratory Standard Operating Procedures.

16.1 The following raw data and records will be retained in the study folder in the study/project archives according to 3M Environmental Laboratory Standard Operating Procedures:

- **16.1.1** Approved protocol and amendments
- **16.1.2** Study correspondence
- **16.1.3** Shipping records
- **16.1.4** Raw data
- **16.1.5** Approved final report (original signed copy)
- **16.1.6** Electronic copies of data

16.2 The following supporting records will be retained separately from the study folder in the archives according to 3M Environmental Laboratory Standard Operating Procedures:

- **16.2.1** Training records
- **16.2.2** Calibration records
- **16.2.3** Instrument maintenance logs
- **16.2.4** Standard Operating Procedures, Equipment Procedures, and Methods

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17. SPECIMEN RETENTION

Specimens will be maintained in the 3M Environmental Laboratory specimen archives for a period of time as specified by regulation or as long as the quality of the preparation affords evaluation, but not longer than ten years following the effective date of the final test rule (if applicable), and as established by 3M Environmental Laboratory Standard Operating Procedures.

18. PROTOCOL AMENDMENTS AND DEVIATIONS

Planned changes to the protocol will be in the form of written amendments signed by the Study Director and the Sponsor's Representative. Amendments will be considered as part of the protocol and will be attached to the final protocol. All changes to the protocol will be indicated in the final report. Any other changes will be in the form of written deviations, signed by the Study Director and filed with the raw data.

19. ATTACHMENTS

19.1 Attachment A Preparatory and analytical methods

20. SIGNATURES

Marvin T Case — 9 June 1999

Marvin T. Case, D.V.M., Ph.D., Sponsor Representative — Date

Kristen Hz — 6/16/99

Kristen J. Hansen, Ph.D., 3M Environmental Laboratory Study Director — Date

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Initial — Date 4/17

Study Title

Oral (Gavage) Pharmacokinetic Study of PFOS in Rats

PROTOCOL AMENDMENT NO. 1

Amendment Date:

August 12, 1999

Performing Laboratory

3M Environmental Technology & Safety Services
3M Environmental Laboratory
935 Bush Avenue
St. Paul, MN 55106

Laboratory Project Identification

ET&SS FACT-TOX110
LIRN U2849

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Date 4/14

This amendment modifies the following portion(s) of the protocol:

1. ***PROTOCOL READS:*** Section 2.0 states this study is designed to determine potassium perfluorooctane sulfonic acid (PFOS) in specimens of liver and serum of rats.

AMEND TO READ: This study is designed to determine PFOS in specimens of rat liver, serum, and urine. All Day -1 and Gestation Day 21 rat urine specimens will be extracted and analyzed by the 3M Environmental Laboratory.

REASON: The urine extraction and analytical methods were not validated and approved prior to protocol approval.

2. ***PROTOCOL READS:*** Section 6.0 lists rat or rabbit liver and serum.

AMEND TO READ: Rat or rabbit urine from 3M Toxicology with a physical description of rat or rabbit urine.

REASON: The rat urine matrix was added after the protocol was approved.

3. ***PROTOCOL READS:*** Section 10.0 and 11.0 list the following methods to use for extraction and analysis:

FACT-M-1.1 "Extraction of Potassium Perfluorooctanesulfonate or Other Anionic Fluorochemical Surfactant from Liver for Analysis Using HPLC-Electrospray/Mass Spectrometry"
FACT-M-2.1 "Analysis of Fluorochemicals in Liver Extracts Using HPLC-Electrospray/Mass Spectrometry"

AMEND TO READ: The extraction and analytical methods to follow at the 3M Environmental Laboratory are:

ETS-8-6.0 "Extraction of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds from Liver for Analysis Using HPLC-Electrospray/Mass Spectrometry"
ETS-8-96.0 "Extraction of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds from Urine for Analysis Using HPLC-Electrospray/Mass Spectrometry/Mass Spectrometry"
ETS-8-7.0 "Analysis of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds in Liver Extracts Using HPLC-Electrospray/Mass Spectrometry"
ETS-8-97.0 "Analysis of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds in Urine Extracts Using HPLC-Electrospray/Mass Spectrometry/Mass Spectrometry"

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REASON: The extraction and analytical methods FACT-M-1.1 and FACT-M-2.1, respectively, were updated on 07/22/99 to ETS-8-6.0 and ETS-8-7.0.
Methods ETS-8-96.0 and ETS-8-97.0 were not validated and approved until after the protocol was approved.

4. ***PROTOCOL READS:*** Section 10.4 and 11.4 state that if the analyses are sub-contracted to other laboratories an amendment will be written to include these methods.

AMEND TO READ: The extraction and analytical methods to follow at Advanced Bioanalytical Services will be attached to the protocol.

The extraction and analytical method to follow at Battelle Memorial Institute is:

"Method for Analysis of Perfluorooctane Sulfonate (PFOS) in Rat ~~Sera~~ (1) Liver by LC/MS/MS, Version ~~1~~ (1) 1.0"

REASON: The analytical methods at the sub-contract laboratories were not included in the original protocol.

5. ***PROTOCOL READS:*** Section 12.2.1 b) Liver method detection limit is 15 ppb.

AMEND TO READ: 12.2.1 b) Liver method detection limit is 8.50 ppb (ng/g).
12.2.1 c) Urine method detection limit is 1.5 ppb (ng/g).

REASON: The validation supporting methods ETS-8-6.0 and ETS-8-7.0 included a lower method detection limit for PFOS in liver. A validation in urine was performed, after protocol approval, to support this method detection limit.

(1) (RE) AC 9/27/99

kjh 9/27/99

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Date 4/15

Amendment Approval

Marvin Case

Marvin Case, D.V.M., Ph.D., Sponsor Representative — 17 August 1999 Date

Kris J. Hansen, Ph.D., Study Director — 8/18/99 Date

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Date 4/19

Study Title

Oral (Gavage) Pharmacokinetic Study of PFOS in Rats

PROTOCOL AMENDMENT NO. 2

Amendment Date:

September 28, 1999

Performing Laboratory

3M Environmental Technology & Safety Services
3M Environmental Laboratory
935 Bush Avenue
St. Paul, MN 55106

Laboratory Project Identification

ET&SS FACT-TOX110
LIRN U2849

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Initial
Date 4/17

This amendment modifies the following portion(s) of the protocol:

1. ***PROTOCOL READS:***
Section 2 states that all liver samples will be extracted and analyzed at Battelle Memorial Institute.

AMEND TO READ:
All dam liver specimens (female mother) and 27 fetus (pooled) liver specimens will be analyzed at Battelle Memorial Institute.

REASON:
The fetal liver/lung is not a target matrix and will not be extracted or analyzed.

2. ***PROTOCOL READS:***
Section 9 states that 80 dam liver specimens (female mother) and 80 fetus (pooled) liver specimens will be sent to the Environmental Laboratory.

AMEND TO READ:
Seventy-three dam liver specimens (female mother), 27 fetus (pooled) liver specimens, and 81 fetal liver/lung specimens (27 liver/lung specimens sampled in triplicate) were sent to the Environmental Laboratory.

REASON:
The number of animals changed during the course of the study.

3. ***PROTOCOL READS:***
Section 12.2.1 a) serum method detection limit is 1.75 ppb b) Liver method detection limit is 15 ppb.

AMEND TO READ:
The method detection limits for all compounds and matrices will be taken from the methods used for extraction and analysis.

REASON:
The method detection limits listed are specific to the 3M Environmental Laboratory. Statement was added to allow for sub-contracted analyses and/or revised methods.

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4. ***PROTOCOL READS:***
Section 16 states that the original data, or copies thereof, will be available at the 3M Environmental Laboratory to facilitate audits of the study during its progress and before acceptance of the final report. When the final report is completed, all original paper data, including: approved protocol and amendments, study correspondence, shipping records, raw data, approved final report, electronic copies of data, training records, calibration records, instrument maintenance logs and standard operating procedures, equipment procedures, and methods will be retained in the archives of the 3M Environmental Laboratory.

AMEND TO READ:
Section 16 states that the original data, or copies thereof, will be available at the 3M Environmental Laboratory to facilitate audits of the study during its progress and before acceptance of the final report. When the final report is completed, all original paper data, including: approved protocol and amendments, study correspondence, shipping records, raw data, approved final report, and electronic copies of data will be retained in the archives of the 3M Environmental Laboratory. All corresponding training records, calibration records, instrument maintenance logs, standard operating procedures, equipment procedures, and methods will be retained in the archives of the facility performing each analysis.

REASON:
Clarification of the disposition of archived records if analyses are performed at a sub-contract laboratory.

5. ***PROTOCOL READS:***
Section 17 states that specimens will be maintained in the 3M Environmental Laboratory specimen archives.

AMEND TO READ:
Specimens will be maintained in the 3M Environmental Laboratory specimen archives. All specimens sent to sub-contract laboratories will be returned to the 3M Environmental Laboratory upon completion of analysis and submission of the sub-contract laboratory(s) final report. The specimens will be returned with the following documentation: the signed original chain of custody and records of storage conditions while at the sub-contract facility.

REASON:
Clarification of the disposition of documentation when shipping specimens for analyses performed by a sub-contract laboratory.

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Amendment Approval

Marvin T Case — 4 Oct 1999

Marvin Case, D.V.M., Ph.D., Sponsor Representative — Date

Kristen Hansen — 5 Oct 1999

Kristen J. Hansen, Ph.D., Study Director — Date

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Date

Study Title

Analytical Laboratory Report on the Determination of Perfluorooctanesulfonate (PFOS) Presence and Concentration in Serum, Liver, and Urine from the Gavage Study of T-6295.12

PROTOCOL AMENDMENT NO. 3

Amendment Date:

20 January 2000

Performing Laboratories

Urine Analyses

3M Environmental Technology and Safety Services
Fluorine Analytical Chemistry Team
Building 2-3E-09
935 Bush Avenue
St. Paul, MN 55106

Liver Analyses

Battelle Memorial Institute
505 King Avenue
Columbus, OH 43201-2693

Serum Analyses

Advanced Bioanalytical Services, Inc.
15 Catherwood Road
Ithaca, NY 14850

Laboratory Project Identification

ET&SS LRN-U2849
FACT TOX-110
Argus Study: 418-013
3M Medical Department Study: T-6295.12

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Date 4/17

This amendment modifies the following portion(s) of the protocol:

1. ***PROTOCOL READS:***
   The study director for the present study was identified in the protocol as Kristen J. Hansen, Ph.D.
   AMEND TO READ:
   The role of study director for the present study was reassigned to Marvin T. Case, D.V.M., Ph.D., as of 20 January 2000. The previous study director, Kristen J. Hansen, has been reassigned to the role of Principle Analytical Investigator.
   REASON:
   The role of study director was reassigned in an effort to ensure compliance with Good Laboratory Practice Standards that outline study personnel requirements (refer to 21 CFR Part 58).

2. ***PROTOCOL READS:***
   The sponsor for the present study was identified as Marvin T. Case, D.V.M., Ph.D.
   AMEND TO READ:
   The role of sponsor for the present study was reassigned to John L. Butenhoff, Ph.D., as of 20 January 2000.
   REASON:
   To ensure that the study director does not also carry the duties of study sponsor, the sponsor role was reassigned. In this manner, personnel responsibilities and workload are more evenly balanced.

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Amendment Approval

John L. Butenhoff — February 10, 2000

John L. Butenhoff, Ph.D., Sponsor Representative — Date

Kristen Hansen — 11-Feb-2000

Kristen J. Hansen, Ph.D., Outgoing Study Director — Date

Marvin T Case — 10 February 2000

Marvin T. Case, D.V.M., Ph.D., Incoming Study Director — Date

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Date

Study Title

Oral (Gavage) Pharmacokinetic Study of PFOS in Rats

PROTOCOL AMENDMENT NO. 4

Amendment Date:

20 April 2000

Performing Laboratories

URINE ANALYSES

3M Environmental Technology and Safety Services
Fluorine Analytical Chemistry Team
Building 2-3E-09, 935 Bush Avenue
St. Paul, MN 55106

FECES ANALYSES

Centre Analytical Laboratories, Inc.
3048 Research Drive
State College, PA 16801

LIVER ANALYSES

Battelle Memorial Institute
505 King Avenue
Columbus, OH 43201-2693

SERUM ANALYSES

Advanced Bioanalytical Services, Inc.
15 Catherwood Road
Ithaca, NY 14850

Laboratory Project Identification

ET&SS LRN-U2849
FACT TOX-110
Argus Study: 418-013
3M Medical Department Study: T-6295.12

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Date 4/19/2000

This amendment modifies the following portion(s) of the protocol:

1. ***PROTOCOL READS:***
   The amended section 2.0 text states that this study is designed to determine PFOS in specimens of rat liver, serum, and urine.
   AMEND TO READ:
   This study is designed to determine PFOS in specimens of rat liver, serum, feces, and urine.
   REASON:
   The analysis of fecal tissue for the target chemical and/or its analytes was added to the scope of the study following the issuance of the protocol. Feces extraction and analytical methods were not validated and approved prior to protocol approval.

2. ***PROTOCOL READS:***
   The amended section 6.0 lists rat or rabbit liver, serum, and urine.
   AMEND TO READ:
   Add: rat or rabbit feces with a physical description of rat or rabbit feces.
   REASON:
   Analysis of fecal tissue for the target chemical and/or its analytes was added to the scope of the study following the issuance of the original protocol.

3. ***PROTOCOL READS:***
   Section 13.1 lists all of the laboratories that will be conducting analyses for this study.
   AMEND TO READ:
   Add: Centre Analytical Laboratories, Inc., 3048 Research Drive, State College, PA 16801
   REASON:
   Feces analyses were added to the scope of this study. The sub-contract laboratory performing analyses was not in the original protocol.

4. ***PROTOCOL READS:***
   Sections 10.4 and 11.4 state that if the analyses are sub-contracted to other laboratories an amendment will be written to include these methods.
   AMEND TO READ:
   The feces extraction and analytical method used by **Centre Analytical Laboratories** will be; 00M–023–003 (Revision 2), "Determination of Fluorochemical Residues in Monkey/Rat Feces by LC/MS/MS."
   REASON:
   The sub-contract laboratory performing feces analyses was added to the scope of this study; this method was not validated and approved prior to protocol approval.

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Amendment Approval

John L. Butenhoff — April 19, 2000

John L. Butenhoff, Ph.D., Sponsor Representative — Date

Marvin T. Case — 19 April 2000

Marvin T. Case, D.V.M., Ph.D., Study Director — Date

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