FINAL REPORT

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

Author
Alan M. Hoberman, Ph.D., DABT, Fellow ATS
(Study Director)

Study Completed On
Final Report 14 May 2010

Sponsor
Daikin Industries, LTD
Chemical Division
Umeda Center Building
4-12 Nakazaki-Nishi, 2-chrome
Kita-ku, Osaka 530-8323
JAPAN

Performing Laboratory
Charles River Laboratories
Preclinical Services
905 Sheehy Drive, Building A
Horsham, PA 19044
USA

Subcontractor Facility
Charles River Laboratories
Preclinical Services
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA

Charles River Laboratories Preclinical Services Protocol Number: UZS00009

STATEMENT OF NO DATA CONFIDENTIALITY CLAIMS

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

This statement supersedes any other claims of confidentiality found in this report.

Company:

Company Agent:

Title:

Date: _____

Signature: _____

1. GLP COMPLIANCE STATEMENT

This final report accurately reflects the raw data obtained during the performance of the study. No deviations from the Good Laboratory Practice (GLP) regulations of the U.S. Environmental Protection Agency [a], the Japanese Ministry of Agriculture, Forestry and Fisheries [b], and the Organisation for Economic Co-operation and Development[c] occurred that affected the quality or integrity of the study, with the following exceptions.

- The extension study was conducted non-GLP, using good scientific practices and according to the Standard Operating Procedures of the Testing Facility.
- The dose formulation analysis and bioanalytical analysis conducted by Charles River Laboratories Preclinical Services Montreal was conducted FDA, OECD and EPA compliant.
- The pharmacokinetic analysis conducted by Charles River Laboratories Preclinical Services Montreal was conducted OECD and EPA compliant.

Submitter:

Date

Sponsor:

---

14 May 2010

Date

Executive Director, Site Operations and Toxicology
Study Director

---

a. U.S. Environmental Protection Agency. Federal Insecticide, Fungicide and Rodenticide Act (FIFRA); Good Laboratory Practice Standards; Final Rule. 40 CFR Part 160.

b. Japanese Ministry of Agriculture, Forestry and Fisheries (1999). Notification on the Good Laboratory Practice (GLP) Standards for Agricultural Chemicals. 11 Nousan No. 6283.

c. Organisation for Economic Co-operation and Development (1998). The Revised OECD Principles of Good Laboratory Practice [C(97)186/Final].

FLAGGING STATEMENT

I have applied the criteria of 40 CFR 158.34 for flagging studies for potential adverse effects to the results of the attached study. This study neither meets nor exceeds any of the applicable criteria.

Company:

Company Agent:

Title:

Date: _____

Signature: _____

2. QUALITY ASSURANCE STATEMENT

Protocol: UZS00009

This study has been inspected by the Quality Assurance Unit to assure conformance with the Good Laboratory Practice (GLP) regulations promulgated by the U.S. Environmental Protection Agency; the Organisation for Economic Co-operation and Development; and the Japanese Ministry of Agriculture, Forestry, and Fisheries.[a] Reports were submitted in accordance with Standard Operating Procedures as follows:

QA INSPECTION DATES

		Date(s) Findings Submitted to:	
Date(s) of Inspection	Phase(s) Inspected	Study Director	Study Director Management
18-19 MAY 2009	Protocol	19 MAY 2009	19 MAY 2009
22 MAY 2009	Amendment 1	22 MAY 2009	22 MAY 2009
02 JUN 2009	Amendment 2	02 JUN 2009	02 JUN 2009
25 JUN 2009	Amendment 3	25 JUN 2009	25 JUN 2009
02 OCT 2009	Amendment 4	02 OCT 2009	02 OCT 2009
09 NOV 2009	Amendment 5	09 NOV 2009	09 NOV 2009
27 MAY 2009	Test Substance Administration	27 MAY 2009	27 MAY 2009
27 MAY 2009	Blood Collection	03 JUN 2009	03 JUN 2009
10 & 12 JUN 2009	Formulations Data	15 JUN 2009	15 JUN 2009
11-12 JUN 2009	In-life Data	15 JUN 2009	15 JUN 2009
25-26 & 29 JUN 2009	Methods	30 JUN 2009	30 JUN 2009
26 JUN 2009	Report Tables	26 JUN 2009	26 JUN 2009
21 JUL 2009	Results	21 JUL 2009	21 JUL 2009

[a]. The Test Facility Quality Assurance Unit did not audit the extension portion of the study.

Date(s) Findings Submitted to:

Date(s) of Inspection	Phase(s) Inspected	Study Director	Study Director Management
21 JUL 2009	Summary	21 JUL 2009	21 JUL 2009
21 JAN 2010	Revised Report	21 JAN 2010	21 JAN 2010
13-14 MAY 2010	Final Report	14 MAY 2010	14 MAY 2010

The QA Statements provided by the following Test Sites have been reviewed.

Test Site(s)	Phase	QA Statement Location
Charles River Preclinical and Clinical Services, Montreal	Dose Formulation Analysis	Appendix 3
Charles River Preclinical and Clinical Services, Montreal	Pharmacokinetic Analysis	Appendix 4
Charles River Preclinical and Clinical Services, Montreal	Bioanalytical Analysis	Appendix 5

The final report has been reviewed to assure that it accurately describes the materials and methods, and the reported results accurately reflect the raw data.

14 May 2010

Auditor Signature/Date

TABLE OF CONTENTS

1. GLP COMPLIANCE STATEMENT 3
2. QUALITY ASSURANCE STATEMENT 5
3. SUMMARY AND CONCLUSION 11
3.1. Purpose 11
3.2. Methods 11
3.3. Results 12
3.4. Conclusion 12
4. DESCRIPTION OF TEST PROCEDURES 13
4.1. Conduct of Study 13
4.1.1. Sponsor 13
4.1.2. Testing Facility 13
4.1.3. Study Number 13
4.1.4. Purpose of the Study 13
4.1.5. Study Design 13
4.1.6. Ownership of the Study 13
4.1.7. Study Monitor 13
4.1.8. Study Director 14
4.1.9. Technical Performance 14
4.1.9.1. Charles River Laboratories Preclinical Services 14
4.1.10. Report Preparation 14
4.1.11. Report Review 14
4.1.12. Date Protocol Signed 14
4.1.13. Dates of Technical Performance 14
4.1.13.1. Main Study 15
4.1.13.2. Extension Study 15
4.1.14. Records Maintained 15
4.2. Test Substance and Vehicle Information 16
4.2.1. Special Handling Instructions 16
4.2.2. Analysis of Activity/Purity 17
4.2.3. Test Substance and Vehicle Preparation and Storage Conditions 17
4.2.4. Analytical Results 17
4.3. Test System 17
4.3.1. Species/Strain 17
4.3.2. Supplier (Source) 17
4.3.3. Sex 17
4.3.4. Rationale for Test System 18
4.3.5. Test System Data 18
4.3.5.1. Main Study 18
4.3.5.2. Extension Study 18
4.3.6. Method of Randomization 18
4.3.6.1. Main Study 18
4.3.6.2. Extension Study 19
4.3.7. System of Identification 19

4.4. Husbandry 19
4.4.1. Research Facility Registration 19
4.4.2. Study Room 19
4.4.3. Housing 19
4.4.4. Light 19
4.4.5. Sanitization 20
4.4.6. Feed 20
4.4.7. Feed Analysis 20
4.4.8. Water 20
4.4.9. Water Analysis 20
4.5. Methods 21
4.5.1. Dosage Administration 21
4.5.2. Rationale for Dosage Selection 21
4.5.3. Route and Rationale for Route of Administration 21
4.5.4. Method and Frequency of Administration 22
4.5.5. Method of Study Performance 22
4.5.6. Scheduled Sacrifice and Blood Collection 22
4.5.6.1. Main Study 22
4.5.6.2. Extension Study 23
4.5.7. Data Collection and Statistical Analyses 23
5. RESULTS 23
5.1. Mortality and Clinical Observations 23
5.2. Body Weights 23
5.3. Pharmacokinetics 23
6. DISCUSSION AND CONCLUSION 24
7. REFERENCES 25

LIST OF TABLES

Table 1. Clinical Observations - Summary....26

Table 2. Body Weights - Summary....27

Table 3. Clinical Observations - Individual Data....28

Table 4. Body Weights - Individual Data....31

LIST OF APPENDICES

APPENDIX 1 - PROTOCOL 34

APPENDIX 2 - CERTIFICATE OF ANALYSIS 84

APPENDIX 3 - ANALYTICAL REPORT 87

APPENDIX 4 - PHARMACOKINETIC ANALYSIS REPORT 117

APPENDIX 5 - BIOANALYTICAL REPORT 143

APPENDIX 6 - ENVIRONMENTAL AND HUSBANDRY REPORTS 178

3. SUMMARY AND CONCLUSION

3.1. Purpose

The study was designed to evaluate the toxicity of acute exposure of Crl:CD1(ICR) female mice to PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid).

The study was extended in order to analyze the test substance levels in the livers of Crl:CD1(ICR) female mice.

3.2. Methods[a]

Sixty three female Crl:CD(1CR) mice were randomly assigned to three dosage groups (Groups I through III), twenty one mice per group. Solutions of the test substance (PFH Ammonium Salt) and vehicle (Reverse osmosis deionized water) were administered orally via gavage once to these mice on Day 1 of study (DS 1) at doses of 35, 175 and 350 mg/kg/day, the dose volume was 5 mL/kg.

An additional six Crl:CD(1CR) (2 per group) mice were assigned to study for a study extension to determine the levels of the test substance in the livers. Extension study mice were administered the test substance once, at the dosage levels described above.

Checks for viability were made at least twice daily. Clinical observations were recorded prior to dosage administration and prior to sacrifice. Body weights were recorded weekly during the acclimation period and once on the day of dosage administration (for main study mice only).

All main study mice were sacrificed [by carbon dioxide asphyxiation] according to the blood sample collection timepoints and discarded without further evaluation.

On the day of dosage administration, blood samples were collected from three mice per group at each timepoint for main study mice. Samples were collected prior to dosage and at approximately 30 minutes, 2, 4, 6, 8 and 24 hours postdosage. Blood was collected from the vena cava after sacrifice. The samples were transferred into un-coated red top tubes and spun in a centrifuge. The resulting serum was transferred into appropriately labeled polypropylene tubes and immediately frozen on dry ice and maintained frozen until shipment for analysis.

Extension study mice were sacrificed two hours (± 10 minutes) after dosage administration, livers were excised for analysis.

---

a. Detailed descriptions of all procedures used in the conduct of this study are provided in the appropriate sections of this report and in APPENDIX 1 (PROTOCOL).

3.3. Results

All mice survived to scheduled sacrifice.

No adverse clinical observations occurred during the study.

3.4. Conclusion

In this acute pharmacokinetic study, mice were administered a single dose at 35, 175 or 350 mg/kg. All mice survived to scheduled sacrifice, and no adverse clinical signs occurred during this study. In the 35 mg/kg dose group, no PFHxA could be quantified after 6 hours. In the 175 and 350 mg/kg dose groups, the mean concentration of PFHxA was below the LLOQ at 24 hours postdosage, but one individual mouse in each group had quantifiable levels at 24 hours.

Based on these results, dosages of 7, 35 and 175 mg/kg/day were selected for the developmental and perinatal/postnatal reproduction toxicity study.

14 May 2010

Date

Executive Director, Site Operations and Toxicology
Study Director

4. DESCRIPTION OF TEST PROCEDURES

4.1. Conduct of Study

4.1.1. Sponsor

Daikin Industries, LTD, Chemical Division, Umeda Center Building, 4-12 Nakazaki-Nishi, 2-chrome, Kita-ku, Osaka 530-8323, JAPAN

4.1.2. Testing Facility

Charles River Laboratories Preclinical Services, 905 Sheehy Drive, Building A, Horsham, PA 19044, USA

4.1.3. Study Number

UZS00009

4.1.4. Purpose of the Study

The study was designed to evaluate the toxicity of acute exposure of Crl:CD1(ICR) female mice to PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid).

The study was extended in order to analyze the test substance levels in the livers of Crl:CD1(ICR) female mice.

4.1.5. Study Design

This study was conducted in compliance with the Good Laboratory Practice (GLP) regulations of the U.S. Environmental Protection Agency[1], the Ministry of Agriculture, Forestry and Fisheries[2] and the Organisation for Economic Co-operation and Development[3]. The bioanalysis and analytical portion of the study were conducted in accordance with the appropriate OECD Principles of GLP (ENV/MC/CHEM(98)17).

4.1.6. Ownership of the Study

The Sponsor owns the study. All raw data, analyses, reports and preserved tissues are the property of the Sponsor.

4.1.7. Study Monitor

(Daikin Industries, Ltd., 1-1 Nishi Hitotsuya, Settsu City, Osaka, 566-8585, JAPAN)

4.1.8. Study Director

, Ph.D., DABT, Fellow ATS (Executive Director, Site Operations and Toxicology)
Address as cited previously for Testing Facility.

4.1.9. Technical Performance

4.1.9.1. Charles River Laboratories Preclinical Services

4.1.9.1.1. Pennsylvania, USA

4.1.9.1.2. Montreal, CANADA

4.1.10. Report Preparation

4.1.11. Report Review

4.1.12. Date Protocol Signed

20 May 2009

4.1.13. Dates of Technical Performance

Mouse Arrival	19 MAY 2009
Experimental Start Date (OECD)	26 MAY 2009
Experimental Start Date (EPA)	27 MAY 2009
Experimental Termination/Completion Date	05 MAR 2010

4.1.13.1. Main Study

Acute Dosage Administration	27 MAY 2009
Blood Sample Collection and Scheduled Sacrifice	27 MAY 2009 - 28 MAY 2009

4.1.13.2. Extension Study

Mice Transferred	09 NOV 2009
Acute Dosage Administration	12 NOV 2009
Sample Collection and Scheduled Sacrifice	12 NOV 2009

4.1.14. Records Maintained

The original report, raw data and reserve samples of the bulk test substance and bulk vehicle are retained in the archives of the Testing Facility. All unused test substance formulations were discarded at the Testing Facility. Backup samples will be discarded at the Testing Facility following issue of the final report. One of the two containers of the bulk test substance was transferred to Charles River Laboratories - Montreal, CANADA for analysis on 23 April 2009 and documented in the raw data. The remaining test substance will be retained at the Testing Facility for future studies and documented in the raw data.

4.2. Test Substance and Vehicle Information

Test Substance Information			
Name	PFH Ammonium Salt (C-1500N)[a]	Description	Colorless liquid
Storage	Room temperature	Supplier	Sponsor

Lot Number	Date Received	Expiration Date
7005	22 APR 2009	31 JUL 2010

Vehicle Information	
R.O. Deionized Water[b]	c

a. Also known as: PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid)
b. R.O. deionized water is an abbreviation used for reverse osmosis membrane processed deionized water.
c. R.O. deionized water is available from a continuous source at the Testing Facility and is maintained at room temperature.

Sampling			
Bulk Test Substance			
Sample Size: 10 mL			
Date Sampled	Date Shipped	Recipient	Shipping Conditions
27 MAY 2009	27 MAY 2009	Montreal, CANADA[a]	Ambient temperature

Bulk Test Substance Reserve		
Sample Size: 5 mL		
Date Sampled	Storage Condition	Date Archived
25 MAY 2009	Room temperature	05 JUN 2009

Bulk Vehicle Reserve		
Sample Size: 5 mL		
Date Sampled	Storage Conditions	Date Archived
25 MAY 2009	Room temperature	05 JUN 2009

Concentration, Homogeneity and Stability[b]				
Sample Size: 2 mL				
Date Sampled	Date Shipped	Recipient	Shipping Conditions	Purpose
26 MAY 2009	27 MAY 2009	Montreal, CANADA[a]	Ambient temperature	Stability, Concentration and Homogeneity

a. Charles River Laboratories - Montreal, CANADA
b. Quadruplicate samples were taken from the top, middle and bottom of each concentration 24 hours or more after preparation, and no more than 24 hours before dosing. Two samples from each quadruplicate set were shipped for analysis; the remaining samples were retained at the Testing Facility as backup samples (room temperature). Stability of the prepared test substance formulations were documented during the study in conjunction with the concentration analysis. Stability was determined for the lowest, intermediate and highest concentrations after storage at room temperature for at least 10 days.

4.2.1. Special Handling Instructions

Gloves, dust-mist/HEPA-filtered mask, appropriate eye protection and protective clothing were worn during formulation preparation and dosage administration.

4.2.2. Analysis of Activity/Purity

The test substance was considered 95% by weight of PFH acid for the purpose of dosage calculations.

The test substance is a marketed product and characterized by its labeling. Information to document or certify the identity, composition, strength and activity/purity of the test substance was provided by the Sponsor to the Testing Facility. A Certificate of Analysis is available in APPENDIX 2. The Certificate of Analysis was not in compliance with the GLPs listed in this report.

Neither the Sponsor nor the Study Director was aware of any potential contaminants likely to have been present in the vehicle that would have interfered with the results of this study.

4.2.3. Test Substance and Vehicle Preparation and Storage Conditions

Solutions of the test substance were prepared once at the Testing Facility and used within 48 hours of preparation. Prepared formulations were stirred continuously for 24 hours prior to dosage administration and stored at room temperature. The vehicle (R.O. water) was available from a continuous source at the Testing Facility and maintained at room temperature.

4.2.4. Analytical Results

All study samples analyzed for concentration were within the acceptance criteria of ±10% of their target values. For homogeneity, the relative standard deviation of the grand mean for all locations was ≤ 5% for all groups. For each group, all stability samples analyzed were within ±10% of the initial concentrations analysis. Results of concentration, homogeneity and stability analyses are available in APPENDIX 3.

4.3. Test System

4.3.1. Species/Strain

Mouse / Crl:CD1(ICR)

4.3.2. Supplier (Source)

Charles River Laboratories, Inc., Kingston, NY, USA

4.3.3. Sex

Female mice.

4.3.4. Rationale for Test System

The Crl:CD1(ICR) mouse was selected as the Test System because: 1) this strain of mouse has been demonstrated to be sensitive to reproductive and developmental toxicants and has been widely used throughout industry for reproductive and developmental toxicity evaluations; 2) historical data and experience exist at the Testing Facility.

4.3.5. Test System Data

4.3.5.1. Main Study

Number of Mice Acclimated	75
Number of Mice Assigned to Study	63
Approximate Date of Birth	19 MAR 2009
Approximate Age at Arrival	62 days
Weight (g) the Day after Arrival	22. 2 - 28.7
Weight (g) at Study Assignment	24.6 - 28.9

4.3.5.2. Extension Study

Number of Mice Acclimated	6
Number of Mice Assigned to Study	6
Weight (g) at Dosage Administration	32.3 - 37.0

4.3.6. Method of Randomization

4.3.6.1. Main Study

Upon arrival, female mice were assigned to individual housing on the basis of computer-generated random units. Female mice were assigned to study groups using a second computer-generated (weight-ordered) randomization procedure on the day before dosing after at least five days of acclimation.

4.3.6.2. Extension Study

Upon transfer from the Testing Facility General Population, female mice were assigned to individual housing on the basis of computer-generated random units. Female mice were assigned to study groups using a second computer-generated (weight-ordered) randomization procedure on the day before dosing.

4.3.7. System of Identification

Mice were permanently identified by tail tattoo according to the Standard Operating Procedures of the Testing Facility. Female mice were given unique permanent identification numbers when assigned to the study based on body weights recorded on the day before dosage administration at the Testing Facility[a].

4.4. Husbandry

4.4.1. Research Facility Registration

USDA Registration No. 14-R-0144 under the Animal Welfare Act, 7 U.S.C. 2131 *et seq*.

4.4.2. Study Room

The study room was maintained under conditions of positive airflow relative to a hallway and independently supplied with a minimum of ten changes per hour of 100% fresh air that had been passed through 99.97% HEPA filters. Room temperature and humidity were monitored constantly throughout the study. Room temperature was targeted at 64°F to 79°F (18°C to 26°C); relative humidity was targeted at 30% to 70%[b].

4.4.3. Housing

Mice were individually housed in stainless steel, wire-bottomed cages. All cage sizes and housing conditions were in compliance with the *Guide for the Care and Use of Laboratory Animals*[(4)].

4.4.4. Light

An automatically controlled 12-hours light:12-hours dark fluorescent light cycle was maintained. Each dark period began at 1900 hours (± 30 minutes).

---

a. Mice body weights were not recorded on the day of arrival, 19 May 2009. This deviation did not adversely affect the outcome or interpretation of the study because the mice were appropriately identified based on body weights recorded on the day before dosage.

b. See APPENDIX 6 (ENVIRONMENTAL AND HUSBANDRY REPORTS).

4.4.5. Sanitization

Cage pan liners were changed as often as necessary. Cages were changed twice.

4.4.6. Feed

Mice were given *ad libitum* access to Certified Rodent Diet® #5002 checkers (PMI® Nutrition International, St. Louis, MO, USA) in individual feeders.

4.4.7. Feed Analysis

Analyses were routinely performed by the feed supplier. No contaminants at levels exceeding the maximum concentration limits for certified feed or deviations from expected nutritional requirements were detected by these analyses. Copies of the results of the feed analyses are available in APPENDIX 6 and the raw data. Neither the Sponsor nor the Study Director was aware of any potential contaminants likely to have been present in the feed that would have interfered with the results of this study.

4.4.8. Water

Local water that had been processed by passage through a reverse osmosis membrane (R.O. water) was available to the mice *ad libitum* from an automatic watering access system and individual water bottles attached to the cages. Chlorine was added to the processed water as a bacteriostat.

4.4.9. Water Analysis

The processed water is analyzed twice annually for possible chemical contamination (Lancaster Laboratories, Lancaster, PA, USA) and monthly for possible bacterial contamination (QC Laboratories, Southampton, PA, USA). Copies of the results of the water analyses are available in APPENDIX 6 and the raw data. Neither the Sponsor nor the Study Director was aware of any potential contaminants likely to have been present in the water that would have interfered with the results of this study.

4.5. Methods

4.5.1. Dosage Administration

Dosage Group	Number of Mice	Dosage (mg/kg/day)	Concentration (mg/mL)	Dosage Volume (mL/kg)	Mouse Number
I	21	35	7	5	3000[a], 3001[b], 3002[c], 1204 - 1221
II	21	175	35	5	1222 - 1242
III	21	350	70	5	1243 - 1263

The test substance was considered 95% by weight of PFH acid for dosage calculations.

a. Mouse 1201 was excluded from study due to insufficient blood volume being collected during the predosage period and replaced with 3000 on 27 May 2009.
b. Mouse 1202 was excluded from study due to insufficient blood volume being collected during the predosage period and replaced with 3001 on 27 May 2009.
c. Mouse 1203 was excluded from study due to insufficient blood volume being collected during the predosage period and replaced with 3002 on 27 May 2009.

4.5.2. Rationale for Dosage Selection

Perfluorobutyrate (PFBA) is a perfluoroalkyl acid (PFAA) found in the environment. Previous studies have indicated developmental toxicity of PFAAs (perfluorooctane sulfonate [PFOS] and perfluorooctanoate [PFOA]). PFBA/NH_4^+ was given to timed-pregnant CD-1 mice by oral gavage daily from gestation day (DG) 1 to 17 at 35, 175 and 350 mg/kg; controls received water. At DG 18, serum levels of PFBA were 3.8, 4.4 and 2.5 μg/mL, respectively, in the three treated groups. PFBA did not significantly affect maternal weight gain, number of implantations, fetal viability, fetus weight, or incidence of fetal malformations. Incidence of full-litter loss was significantly greater in the 350 mg/kg group, and maternal liver weights were significantly increased in the 175 and 350 mg/kg groups. In contrast to PFOA, and PFOS, PFBA exposure during pregnancy did not adversely affect neonatal survival or postnatal growth. Liver enlargement was detected in the PFBA-exposed pups on postnatal (PD) 1, but not by PD 10. Expression of selected hepatic genes in PFBA-exposed pups at PD 1 did not reveal any significant changes from controls. A significant delay in eye-opening in offspring was detected in all three PFBA groups, and slight delays in the onset of puberty were noted in the 175 and 350 mg/kg groups. These data suggest that exposure to PFBA during pregnancy in the mouse did not produce developmental toxicity comparable to that observed with PFOA, in part, due to rapid elimination of the chemical[(5)]. Based on the results of this study, dosages of 35, 175 and 350 mg/kg/day were selected for the acute pharmacokinetic study.

4.5.3. Route and Rationale for Route of Administration

The oral (gavage) route was selected for use because: 1) in comparison with the dietary route, the exact dosage can be accurately administered; and 2) it is one possible route of human exposure.

4.5.4. Method and Frequency of Administration

Female mice were given the test substance and/or the vehicle once and the dosage was adjusted for the weight on that day.

4.5.5. Method of Study Performance

Mice were observed for viability at least twice daily and for clinical observations and general appearance weekly during the acclimation period. The mice were also examined for clinical observations prior to dosage administration and prior to sacrifice. Body weights were recorded weekly during the acclimation period and once on the day of dosage administration (for main study mice only).

4.5.6. Scheduled Sacrifice and Blood Collection

4.5.6.1. Main Study

Mice were sacrificed [by carbon dioxide asphyxiation] according to the blood sample collection timepoints and discarded without further evaluation.

On the day of dosage administration, blood samples (0.3 mL to 0.5 mL[a]) were collected from three mice per group at each timepoint. Samples were collected prior to dosage and at approximately 30 minutes, 2 hours, 4 hours, 6 hours, 8 hours and 24 hours postdosage. The time of each blood collection was recorded in the raw data.

Blood was collected from the vena cava after sacrifice. The samples were transferred into un-coated red top tubes and spun in a 4^{o}C centrifuge for 10 minutes at 3500 RPM. The resulting serum was transferred into polypropylene tubes labeled at minimum with the protocol number, mouse number, group number, dosage level, day of study, collection interval, date of collection, species, generation and storage conditions. All samples were immediately frozen on dry ice and maintained frozen (≤-60°C) until shipment for analysis (on dry ice) to Charles River Laboratories - Montreal, CANADA. Results of the pharmacokinetic and bioanalytical analysis are available in APPENDICES 4 and 5.

---

a. During the 2 hour postdosage period, mouse 1208 (Group I) had 0.3 mL of blood collected; during the 4 hour postdosage period, mouse 1233 (Group II) had 0.3 mL of blood collected; during the 8 hour postdosage period, mouse 1259 (Group III) had 0.4 mL of blood collected; during the 4 hour postdosage period, mouse 1211 (Group I) had 0.4 mL of blood collected; and during the 4 hour postdosage period, mouse 1252 (Group III) had 0.4 mL of blood collected. These deviations did not adversely affect the outcome or interpretation of the study because sufficient blood was collected for analysis.

4.5.6.2. Extension Study

Two hours (± 10 minutes) after dosage administration [mice were sacrificed by carbon dioxide asphyxiation], livers were excised and retained frozen (≤ -70°C) from all mice after sacrifice. The time of each liver collection was documented in the raw data.

Liver samples were shipped (on dry ice) to Charles River Laboratories Preclinical Services, Montreal.

Liver samples were analyzed using a non-validated LC-MS/MS for determination of suitable analytical range.

4.5.7. Data Collection and Statistical Analyses

Averages were calculated.

Data generated during the course of this study were recorded either by hand or using the *Argus Automated Data Collection and Management System* and the *Vivarium Temperature and Relative Humidity Monitoring System*. All data were tabulated and summarized using the *Argus Automated Data Collection and Management System*, the *Vivarium Temperature and Relative Humidity Monitoring System* and Quattro Pro 8.

5. RESULTS

5.1. Mortality and Clinical Observations (Summary - Table 1; Individual Data - Table 3)

All mice survived until scheduled sacrifice.

No adverse clinical observations occurred during the study.

5.2. Body Weights (Summary - Table 2; Individual Data - Table 4)

Body weights on day 1 of study (DS 1) were comparable among the three dosage groups.

5.3. Pharmacokinetics (APPENDIX 4)

The serum concentrations of PFHxA were not quantifiable in predosage samples in any of the groups. In the 35 mg/kg dose group, no PFHxA could be quantified after 6 hours. In the 175 and 350 mg/kg dose groups, the mean concentration of PFHxA was below the LLOQ at 24 hours postdosage, but one individual mouse in each group had quantifiable levels at 24 hours.

6. DISCUSSION AND CONCLUSION

Perfluorobutyrate (PFBA) is a perfluoroalkyl acid (PFAA) found in the environment. Previous studies have indicated developmental toxicity of PFAAs (perfluorooctane sulfonate [PFOS] and perfluorooctanoate [PFOA]). PFBA/NH_4^+ was given to timed-pregnant CD-1 mice by oral gavage daily from gestation day (DG) 1 to 17 at 35, 175 and 350 mg/kg; controls received water. At DG 18, serum levels of PFBA were 3.8, 4.4 and 2.5 μg/mL, respectively, in the three treated groups. PFBA did not significantly affect maternal weight gain, number of implantations, fetal viability, fetus weight, or incidence of fetal malformations. Incidence of full-litter loss was significantly greater in the 350 mg/kg group, and maternal liver weights were significantly increased in the 175 and 350 mg/kg groups. In contrast to PFOA, and PFOS, PFBA exposure during pregnancy did not adversely affect neonatal survival or postnatal growth. Liver enlargement was detected in the PFBA-exposed pups on postnatal (PD) 1, but not by PD 10. Expression of selected hepatic genes in PFBA-exposed pups at PD 1 did not reveal any significant changes from controls. A significant delay in eye-opening in offspring was detected in all three PFBA groups, and slight delays in the onset of puberty were noted in the 175 and 350 mg/kg groups. These data suggest that exposure to PFBA during pregnancy in the mouse did not produce developmental toxicity comparable to that observed with PFOA, in part, due to rapid elimination of the chemical[5,6].

In this acute pharmacokinetic study, mice were administered a single dose at 35, 175 or 350 mg/kg. All mice survived to scheduled sacrifice, and no adverse clinical signs occurred during this study. In the 35 mg/kg dose group, no PFHxA could be quantified after 6 hours. In the 175 and 350 mg/kg dose groups, the mean concentration of PFHxA was below the LLOQ at 24 hours postdosage, but one individual mouse in each group had quantifiable levels at 24 hours.

Based on these results, dosages of 7, 35 and 175 mg/kg/day were selected for the developmental and perinatal/postnatal reproduction toxicity study.

7. REFERENCES

(1) Federal Insecticide, Fungicide and Rodenticide Act/Toxic Substances Control Act (FIFRA/TSCA); Good laboratory practice standards; Final Rule 40 C.F.R Part 160/792; August 17, 1989. U.S. Environmental Protection Agency.

(2) Good laboratory practice standards for toxicological studies on agricultural chemicals. 59-Nousan-No.3850; August 10, 1984. Repealed as 1 October, 1999. Notification 11-Nousan-No.6283. Japan: Ministry of Agriculture, Forestry and Fisheries, Japan (MAFF).

(3) OECD Principles of good laboratory practices, [C(97)186/Final] (1998); Environmental Health and Safety Division. OECD Environment Directorate.

(4) Institute of Laboratory Animal Resources Commission on Life Sciences and the National Research Council. *Guide for the care and use of laboratory animals*. Washington (D.C.): National Academy Press; 1996.

(5) Dar, K., Grey, B., Zehr, B., Wood, C., Butenhoff, J., Chang, S., Ehresman, D., Tan, Y. & Lau, C. 2008, Effects of Perfluorobutyrate Exposure during Pregnancy in the Mouse, Toxicological Sciences 105(1), 173-181 (2008).

(6) Das KP, Grey BE, Zehr RD et al. Effects of perfluorobutyrate exposure during pregnancy in the mouse. *Toxicol Sci* 2008;105(1):173-81.

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 1 (PAGE 1): CLINICAL OBSERVATIONS - SUMMARY

DOSAGE GROUP	I	II	III
DOSAGE (MG/KG/DAY)	35	175	350
MAXIMUM POSSIBLE INCIDENCE	24/ 21	24/ 21	24/ 21
NO ADVERSE FINDINGS			

MAXIMUM POSSIBLE INCIDENCE = (DAYS x MICE)/NUMBER OF MICE EXAMINED PER GROUP

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 2 (PAGE 1): BODY WEIGHTS - SUMMARY

DOSAGE GROUP		I	II	III
DOSAGE (MG/KG/DAY)		35	175	350
MICE TESTED	N	21	21	21
BODY WEIGHT (G)				
DAY 1	MEAN±S.D.	26.7 ± 1.3	26.5 ± 1.0	26.4 ± 1.2

DAY = DAY OF STUDY

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 3 (PAGE 1): CLINICAL OBSERVATIONS - INDIVIDUAL DATA

DOSAGE GROUP I	LOW DOSAGE	35 MG/KG/DAY
MOUSE #	DESCRIPTION	
3000	NO ADVERSE FINDINGS	
3001	NO ADVERSE FINDINGS	
3002	NO ADVERSE FINDINGS	
1204	NO ADVERSE FINDINGS	
1205	NO ADVERSE FINDINGS	
1206	NO ADVERSE FINDINGS	
1207	NO ADVERSE FINDINGS	
1208	NO ADVERSE FINDINGS	
1209	NO ADVERSE FINDINGS	
1210	NO ADVERSE FINDINGS	
1211	NO ADVERSE FINDINGS	
1212	NO ADVERSE FINDINGS	
1213	NO ADVERSE FINDINGS	
1214	NO ADVERSE FINDINGS	
1215	NO ADVERSE FINDINGS	
1216	NO ADVERSE FINDINGS	
1217	NO ADVERSE FINDINGS	
1218	NO ADVERSE FINDINGS	
1219	NO ADVERSE FINDINGS	
1220	NO ADVERSE FINDINGS	
1221	NO ADVERSE FINDINGS	

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 3 (PAGE 2): CLINICAL OBSERVATIONS - INDIVIDUAL DATA

DOSAGE GROUP II	MIDDLE DOSAGE	175 MG/KG/DAY
MOUSE #	DESCRIPTION	
1222	NO ADVERSE FINDINGS	
1223	NO ADVERSE FINDINGS	
1224	NO ADVERSE FINDINGS	
1225	NO ADVERSE FINDINGS	
1226	NO ADVERSE FINDINGS	
1227	NO ADVERSE FINDINGS	
1228	NO ADVERSE FINDINGS	
1229	NO ADVERSE FINDINGS	
1230	NO ADVERSE FINDINGS	
1231	NO ADVERSE FINDINGS	
1232	NO ADVERSE FINDINGS	
1233	NO ADVERSE FINDINGS	
1234	NO ADVERSE FINDINGS	
1235	NO ADVERSE FINDINGS	
1236	NO ADVERSE FINDINGS	
1237	NO ADVERSE FINDINGS	
1238	NO ADVERSE FINDINGS	
1239	NO ADVERSE FINDINGS	
1240	NO ADVERSE FINDINGS	
1241	NO ADVERSE FINDINGS	
1242	NO ADVERSE FINDINGS	

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 3 (PAGE 3): CLINICAL OBSERVATIONS - INDIVIDUAL DATA

DOSAGE GROUP III	HIGH DOSAGE	350 MG/KG/DAY
MOUSE #	DESCRIPTION	
1243	NO ADVERSE FINDINGS	
1244	NO ADVERSE FINDINGS	
1245	NO ADVERSE FINDINGS	
1246	NO ADVERSE FINDINGS	
1247	NO ADVERSE FINDINGS	
1248	NO ADVERSE FINDINGS	
1249	NO ADVERSE FINDINGS	
1250	NO ADVERSE FINDINGS	
1251	NO ADVERSE FINDINGS	
1252	NO ADVERSE FINDINGS	
1253	NO ADVERSE FINDINGS	
1254	NO ADVERSE FINDINGS	
1255	NO ADVERSE FINDINGS	
1256	NO ADVERSE FINDINGS	
1257	NO ADVERSE FINDINGS	
1258	NO ADVERSE FINDINGS	
1259	NO ADVERSE FINDINGS	
1260	NO ADVERSE FINDINGS	
1261	NO ADVERSE FINDINGS	
1262	NO ADVERSE FINDINGS	
1263	NO ADVERSE FINDINGS	

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 4 (PAGE 1): BODY WEIGHTS - INDIVIDUAL DATA

MOUSE #	DOSAGE GROUP I	LOW DOSAGE	35 MG/KG/DAY
	DAY 1		
3000	24.0		
3001	29.0		
3002	24.0		
1204	26.5		
1205	25.2		
1206	28.3		
1207	26.5		
1208	26.3		
1209	25.1		
1210	28.0		
1211	26.5		
1212	27.0		
1213	25.8		
1214	27.9		
1215	26.2		
1216	26.7		
1217	27.6		
1218	27.5		
1219	26.9		
1220	27.4		
1221	27.5		

DAY = DAY OF STUDY
ALL WEIGHTS WERE RECORDED IN GRAMS (G).

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 4 (PAGE 2): BODY WEIGHTS - INDIVIDUAL DATA

MOUSE #	DOSAGE GROUP II	MIDDLE DOSAGE	175 MG/KG/DAY
	DAY 1		
1222	25.0		
1223	29.5		
1224	26.9		
1225	26.0		
1226	26.6		
1227	27.2		
1228	26.4		
1229	25.5		
1230	25.2		
1231	27.9		
1232	26.2		
1233	26.4		
1234	26.0		
1235	26.8		
1236	26.8		
1237	26.8		
1238	26.1		
1239	27.2		
1240	25.6		
1241	26.2		
1242	25.7		

DAY = DAY OF STUDY
ALL WEIGHTS WERE RECORDED IN GRAMS (G).

PROTOCOL UZS00009: ORAL (GAVAGE) ACUTE PHARMACOKINETIC STUDY OF PFH AMMONIUM SALT (AMMONIUM SALT OF PERFLOURINATED HEXANOIC ACID) IN MICE

TABLE 4 (PAGE 3): BODY WEIGHTS - INDIVIDUAL DATA

MOUSE #	DOSAGE GROUP III	HIGH DOSAGE	350 MG/KG/DAY
	DAY 1		
1243	25.0		
1244	27.6		
1245	26.9		
1246	26.7		
1247	24.0		
1248	28.0		
1249	25.7		
1250	25.9		
1251	24.9		
1252	27.0		
1253	27.0		
1254	26.8		
1255	24.4		
1256	27.7		
1257	27.0		
1258	26.8		
1259	25.6		
1260	27.6		
1261	25.2		
1262	27.3		
1263	26.6		

DAY = DAY OF STUDY
ALL WEIGHTS WERE RECORDED IN GRAMS (G).

APPENDIX 1 - PROTOCOL

charles river

FINAL PROTOCOL

Charles River Laboratories Study No. UZS00009

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

SPONSOR:
Daikin Industries, LTD
Chemical Division
Umeda Center Building
4-12 Nakazaki-Nishi, 2-chrome
Kita-ku, Osaka 530-8323
JAPAN

PERFORMING LABORATORY:
Charles River Laboratories
Preclinical Services
905 Sheehy Drive, Building A
Horsham, PA 19044
USA

20 May 2009

TABLE OF CONTENTS

1. STUDY NUMBER 4
2. STUDY TITLE 4
3. PURPOSE 4
4. TESTING FACILITY 4
5. STUDY DIRECTOR 4
6. SPONSOR 4
7. STUDY MONITOR 5
8. DOSE FORMULATION ANALYSIS SCIENTIST 5
9. PHARMACOKINETIC ANALYSIS SCIENTIST 5
10. TOXICOKINETIC ANALYSIS SCIENTIST 5
11. REGULATORY COMPLIANCE 6
12. PROPOSED STUDY SCHEDULE 6
13. TEST SUBSTANCE AND VEHICLE 7
14. FORMULATION 8
15. ANALYSES 8
16. DISPOSITION 10
17. TEST SYSTEM 10
18. ANIMAL HUSBANDRY 11
19. RANDOMIZATION 12
20. ADMINISTRATION 13
21. TESTS, ANALYSES AND MEASUREMENTS 14
22. METHOD OF SACRIFICE 16

23. PROPOSED STATISTICAL METHODS 17

24. DATA ACQUISITION, VERIFICATION AND STORAGE 17

25. RECORDS TO BE MAINTAINED 18

26. KEY PERSONNEL 18

27. FINAL REPORT 19

28. ANIMAL WELFARE 19

29. INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE STATEMENT 20

30. REFERENCES 21

31. PROTOCOL APPROVAL 22

ATTACHMENT 1 - PROPOSED STUDY SCHEDULE 24

ATTACHMENT 2 - MATERIAL SAFETY DATA SHEET 26

ATTACHMENT 3 - TEST SUBSTANCE PREPARATION PROCEDURE 32

1. STUDY NUMBER

UZS00009

2. STUDY TITLE

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

3. PURPOSE

The purpose of this study is to evaluate the toxicity of acute exposure of Crl:CD1(ICR) female mice to PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid).

4. TESTING FACILITY

Charles River Laboratories
Preclinical Services
905 Sheehy Drive, Building A
Horsham, PA 19044
USA
Main Tel: 215.443.8710
Fax: 215.443.8587

5. STUDY DIRECTOR

Address as cited for Testing Facility.
E-mail:

6. SPONSOR

Daikin Industries, LTD
Chemical Division
Umeda Center Building
4-12 Nakazaki-Nishi, 2-chrome
Kita-ku, Osaka 530-8323
JAPAN

7. STUDY MONITOR

Daikin Industries, Ltd.
1-1 Nishi Hitotsuya
Settsu City
Osaka, 566-8585
JAPAN
Tel: +81.6.6349.5336
Fax: +81.6.6349.1095
E-mail:

8. DOSE FORMULATION ANALYSIS SCIENTIST

Principal Investigator:
Research Scientist, Analytical Chemistry
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA
Tel: +1.514.630.8200 ext. 2561
Fax: +1.514.630.8200
E-mail:

9. PHARMACOKINETIC ANALYSIS SCIENTIST

To be added by amendment

10. TOXICOKINETIC ANALYSIS SCIENTIST

Principal Investigator: PharmD, Ph.D.
Senior Research Scientist, Pharmacokinetics
Charles River Preclinical and Clinical Services
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA
Tel: +1.514.630.8200, ext. 8368
Fax: +1.514.630.8230
E-mail:

11. REGULATORY COMPLIANCE

This study will be conducted in compliance with the Good Laboratory Practice (GLP) regulations of the U.S. Environmental Protection Agency[1], the Ministry of Agriculture, Forestry and Fisheries[2] and the Organisation for Economic Co-operation and Development[3].

All changes or revisions of this protocol shall be documented, approved by the Institutional Animal Care and Use Committee, signed by the Study Director, dated and maintained with the protocol.

The Testing Facility's Quality Assurance Unit (QAU) will audit the protocol, the raw data and the report, and will inspect critical phases of those portions of the study conducted at the Testing Facility in accordance with the Standard Operating Procedures of the Testing Facility.

The final report will include a compliance statement signed by the Study Director that the report accurately reflects the raw data obtained during the performance of the study and that all applicable GLP regulations were followed in the conduct of the study. Should deviations from GLP regulations occur, each will be described in detail, together with how the deviation might affect the quality or integrity of the study.

Should any portion of the study be conducted by a subcontractor or by the Sponsor, the Testing Facility management will ensure that a qualified Principal Investigator is identified by the site conducting that portion of the study. All procedures conducted by the Test Site will be specifically defined by the protocol, or will be described in detail in the Standard Operating Procedures of the Test Site. The QAU for this facility site will conduct critical phase inspections and audit respective results and reports for that study portion according to the SOPs of that site. Such critical phase inspection reports and report audits will be submitted by the site to the Principal Investigator and the Study Director. The dates of the inspections and report submissions will be incorporated into a QAU Statement generated by that site and provided to the Testing Facility for inclusion in the final report. In addition, this site will provide a statement of GLP compliance, as described above, signed by the Principal Investigator for inclusion in the final report. The archival location of any records generated by this site will be identified in the final report.

12. PROPOSED STUDY SCHEDULE

See Attachment 1 to the Protocol.

13. TEST SUBSTANCE AND VEHICLE

13.1. Identification

13.1.1. Test Substance

PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid). Ammonium Perfluorohexanoate's CAS number is: 21615-47-4.

The test substance will be supplied as a 50% aqueous solution (lot identification will be documented in the raw data).

The Sponsor will provide to the Testing Facility documentation or certification of the identity, composition, strength and activity/purity of the test substance. This documentation will be included in the final report. The Sponsor's signature and approval of the protocol indicates that appropriate documentation of the method of synthesis, fabrication or derivation of the test and control substances is on file and that it is available to the appropriate regulatory agencies should it be requested.

13.1.2. Vehicle

Reverse osmosis membrane processed deionized water (R.O. deionized water). There will be no lot number for R.O. deionized water; this material is available from a continuous source at the Testing Facility.

Neither the Sponsor nor the Study Director is aware of any potential contaminants likely to be present in the vehicle that would interfere with the results of this study. Therefore, no analyses other than those mentioned in this protocol will be conducted.

13.2. Safety Precautions

Double nitrile gloves, dust-mist/HEPA-filtered mask, appropriate eye protection, uniform/lab coat and sleeves to be worn during formulation preparation and dosage. The Material Safety Data Sheet (MSDS) is attached to the protocol (Attachment 2).

13.3. Storage

Bulk Test Substance:	Room temperature
Vehicle:	Room temperature
Prepared Formulations:	Room temperature

All test substance shipments should be addressed to the attention of Mark Coker (mark.coker@crl.com), Manager of Formulation Laboratory, at the previously cited Testing Facility address and telephone number.

Shipments should include information concerning storage conditions and shipping cartons should be labeled appropriately. The recipient should be notified in advance of shipment.

14. FORMULATION

14.1. Frequency of Preparation

Formulations (solutions) will be prepared at least once at the Testing Facility and used within 48 hours of preparation. Prepared formulations will be stirred continuously for **24 hours** prior to dosage administration.

Detailed preparation procedures are attached to this protocol (Attachment 3).

14.2. Adjustment for Activity/Purity

The test substance will be considered 95% by weight of PFH acid for dosage calculations.

14.3. Testing Facility Reserve Samples

The Testing Facility will reserve a sample of 5 mL of each lot of bulk test substance and bulk vehicle used during the course of the study. Samples will be stored under the previously cited conditions.

15. ANALYSES

The Sponsor will provide to the Testing Facility documentation or certification of the identity, composition, strength and activity/purity of the test substance. Results of these analyses will be included in the study report.

Samples additional to those described below may be taken if deemed necessary during the course of the study. Additional analyses, if required, will be documented by protocol amendment.

15.1. Acceptance Criteria

Acceptance criteria for analytical results for each group are defined as follows: 1) concentration results will be considered acceptable if the difference between the actual mean value and the targeted concentration is ± 10%; 2) homogeneity results for a group will be considered acceptable if the relative standard deviation (RSD) for the formulation, calculated as the RSD for the grand mean of the average values for top, middle and bottom locations, is ≤5%; and 3) results of 10 day stability analysis will be within ± 10% of the concentration of the initial stability analysis. Results obtained outside of the criteria will be considered Out of Specification (OOS) and procedures for investigation and notification will be followed in the applicable laboratory Standard Operating Procedure covering OOS results.

15.2. Bulk Test Substance Stability

A sample of approximately 10 mL of the test substance will be taken on the last day of treatment and sent (ambient conditions) for analysis.

15.3. Analyses of Prepared Formulations

15.3.1. Concentration and Homogeneity

Concentration and homogeneity of the prepared formulations will be verified during the course of this study. Quadruplicate samples (2 mL each) will be taken from the top, middle and bottom of each concentration 24 hours or more after preparation, and no more than 24 hours before dosing. Two samples from each quadruplicate set will be shipped (ambient conditions) for analysis; the remaining samples will be retained at the Testing Facility as backup samples. The average of the homogeneity results will be reported as the concentration values for the preparation. Backup samples will be stored under the previously cited bulk test substance storage conditions (room temperature).

15.3.2. Stability

Stability of the prepared test substance formulations will be documented during this study in conjunction with a concentration analysis. Stability will be determined for the lowest and highest concentrations after storage at room temperature for at least 10 days.

15.4. Shipping Instructions

This sample will be sent (ambient conditions) to:

Principal Investigator: BSc
Research Scientist, Analytical Chemistry
Charles River Preclinical and Clinical Services
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA
Tel: +1.514.630.8200 ext 2516
Fax: +1.514.630.8200
E-mail:

The recipient will be notified in advance of sample shipment.

16. DISPOSITION

Unused prepared formulations will be discarded at the Testing Facility. Backup samples will be discarded at the Testing Facility following issue of the final report. Disposition of the remaining bulk test substance will be documented in the raw data.

17. TEST SYSTEM

17.1. Species/Strain and Reason for Selection

The Crl:CD1(ICR) mouse was selected as the Test System because: 1) this strain of mouse has been demonstrated to be sensitive to reproductive and developmental toxicants and has been widely used throughout industry for reproductive and developmental toxicity evaluations; 2) historical data and experience exist at the Testing Facility.

17.2. Number

Initial population acclimated: 75 female mice.
Population selected for study: 63 female mice (21 per dosage group).

17.3. Sex

Female mice.

17.4. Body Weight and Age

Female mice will be ordered to be approximately 60 days of age at receipt, at which time they will be expected to have body weights of 25 g to 30 g each. Actual body weights will be recorded the day after receipt and will be documented in the raw data. The weight range will be included in the final report.

17.5. Source

Charles River Laboratories, Inc.

The mice will be shipped in filtered cartons by air freight and/or truck from Charles River Laboratories, Inc., to the Testing Facility.

17.6. Identification

Mice are permanently identified by tail tattoo according to the Standard Operating Procedures of the Testing Facility. Female mice will be given unique permanent identification numbers when assigned to the study based on body weights recorded on the day of arrival at the Testing Facility.

18. ANIMAL HUSBANDRY

All cage sizes and housing conditions are in compliance with the *Guide for the Care and Use of Laboratory Animals*[4].

18.1. Housing

The mice will be individually housed in stainless steel, wire-bottomed cages.

18.2. Room Air, Temperature and Humidity

The animal room is independently supplied with at least ten changes per hour of 100% fresh air that has been passed through 99.97% HEPA filters. Room temperature will be maintained at 64°F to 79°F (18°C to 26°C) and monitored constantly. Room humidity will also be monitored constantly and maintained at 30% to 70%.

18.3. Light

An automatically controlled 12-hour light:12-hour dark fluorescent light cycle will be maintained. Each dark period will begin at 1900 hours (± 30 minutes). The light cycle may be adjusted by the Study Director or designee if deemed necessary to accommodate scheduled laboratory activities. Any such adjustment will be documented in the raw data.

18.4. Feed

Mice will be given Certified Rodent Diet® #5002 checkers (PMI® Nutrition International) available *ad libitum* from individual feeders.

18.5. Water

Water will be available *ad libitum* from individual bottles attached to the cages and/or from an automatic watering access system. All water will be from a local source and passed through a reverse osmosis membrane before use. Chlorine will be added to the processed water as a bacteriostat; processed water is expected to contain no more than 1.2 ppm chlorine at the time of analysis. Water is analyzed monthly for possible bacterial contamination and twice annually for possible chemical contamination.

18.6. Contaminants

Neither the Sponsor nor the Study Director is aware of any potential contaminants likely to be present in the certified diet or the drinking water at levels that would interfere with the results of this study. Therefore, no analyses other than those routinely performed by the feed supplier or those mentioned in this protocol will be conducted.

19. RANDOMIZATION

Upon arrival, female mice will be assigned to individual housing on the basis of computer-generated random units. Female mice will be assigned to study groups using a second computer-generated (weight-ordered) randomization procedure on the day before dosing after at least five days of acclimation (before dosing).

20. ADMINISTRATION

20.1. Route and Reason for Choice

The oral (gavage) route was selected for use because: 1) in comparison with the dietary route, the exact dosage can be accurately administered; and 2) it is one possible route of human exposure.

20.2. Method and Frequency

Female mice will be given the test substance and/or the vehicle once and the dosage will be adjusted for the weight on that day.

20.3. Rationale for Dosage Selection

Perfluorobutyrate (PFBA) is a perfluoroalkyl acid (PFAA) found in the environment. Previous studies have indicated developmental toxicity of PFAAs (perfluorooctane sulfonate [PFOS] and perfluorooctanoate [PFOA]). PFBA/NH_4^+ was given to timed-pregnant CD-1 mice by oral gavage daily from gestation day (DG) 1 to 17 at 35, 175 and 350 mg/kg; controls received water. At DG 18, serum levels of PFBA were 3.8, 4.4 and 2.5 μg/mL, respectively, in the three treated groups. PFBA did not significantly affect maternal weight gain, number of implantations, fetal viability, fetus weight, or incidence of fetal malformations. Incidence of full-litter loss was significantly greater in the 350 mg/kg group, and maternal liver weights were significantly increased in the 175 and 350 mg/kg groups. In contrast to PFOA, and PFOS, PFBA exposure during pregnancy did not adversely affect neonatal survival or postnatal growth. Liver enlargement was detected in the PFBA-exposed pups on postnatal (PD) 1, but not by PD 10. Expression of selected hepatic genes in PFBA-exposed pups at PD 1 did not reveal any significant changes from controls. A significant delay in eye-opening in offspring was detected in all three PFBA groups, and slight delays in the onset of puberty were noted in the 175 and 350 mg/kg groups. These data suggest that exposure to PFBA during pregnancy in the mouse did not produce developmental toxicity comparable to that observed with PFOA, in part, due to rapid elimination of the chemical[5]. Based on the results of this study, dosages of 35, 175 and 350 mg/kg/day were selected for the acute pharmacokinetic study.

20.4. Dosage Levels, Concentrations and Dosage Volumes

Dosage Group	Number of Mice	Dosage (mg/kg/day)	Concentration (mg/mL)	Dosage Volume (mL/kg)	Batch Number
I	21	35	7	5	B-UZS00009-A(Day.Month.Year)
II	21	175	35	5	B-UZS00009-B(Day.Month.Year)
III	21	350	70	5	B-UZS00009-C(Day.Month.Year)

The test substance will be considered 95% by weight of PFH acid for dosage calculations.

21. TESTS, ANALYSES AND MEASUREMENTS

21.1. Viability

All Periods: At least twice daily.

21.2. Clinical Observations and/or General Appearance

Acclimation Period: At least weekly.

Dosage Period: Prior to dosage administration. Postdosage observations will be recorded prior to sacrifice for blood collection.

Clinical observations may be recorded more frequently than cited above.

21.3. Body Weights

Acclimation Period: At least weekly.

Dosage Period: Once on the day of dosage administration.

21.4. Feed Consumption Values

Feed consumption values will not be recorded during this study. Feed consumption will be monitored and feed will be replenished on an as-needed basis.

21.5. Pharmacokinetic Sample Collection

On the day of dosage administration, blood samples (0.5 mL each ± 15%) will be collected from three mice per group at each timepoint. Samples will be prior to dosage and at approximately 30 minutes, 2 hours, 4 hours, 6 hours, 8 hours and 24 hours postdosage. The time of each blood collection will be recorded in the raw data.

Blood will be collected from the vena cava after sacrifice. (If necessary, blood may be collected from an alternate site; if so, the alternate site will be documented in the raw data.) The samples will be transferred into un-coated red top tubes and spun in a centrifuge. The resulting serum will be transferred into polypropylene tubes labeled at minimum with the protocol number, mouse number, group number, dosage level, day of study, collection interval, date of collection, species, generation and storage conditions. All samples will be frozen on dry ice as soon as possible and maintained frozen (≤60°C) until shipment for analysis.

21.5.1. Shipping Instructions

Samples to be analyzed will be shipped (on dry ice) to:

Principal Investigator: To be added by amendment
ATT:
Charles River Preclinical and Clinical Services
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA
Custom Clearance: H. Kennedy Inc
Tel: +1.514.630.8200 ext 8974
Fax: +1.514.630.8200
E-mail:

The recipient will be notified in advance of sample shipment. Copies of blood collection data sheets will be included in the shipment.

21.6. Pharmacokinetic Sample Analysis

To be added by amendment.

21.7. Toxicokinetic Data Analysis

The toxicokinetic (TK) profile of each group will be characterized by non-compartmental analysis of the test article serum concentration data using validated computer software (WinNonlin, version 3.2, Pharsight Corp., Mountain View, California, USA). The area under the test article serum concentration vs. time curve (AUC) will be calculated using the linear trapezoidal method (linear interpolation). If possible, the terminal elimination phase of the TK profile will be identified and its slope calculated using log-linear regression. The coefficient of determination of the line fitted to the terminal elimination phase will be calculated. TK parameters describing the systemic exposure of the test substance in the test system will be estimated from observed serum concentration values, the dosing regimen, the AUC, and the terminal elimination phase rate constant for each group. Analysis will be performed under Project Number 480173. Descriptive statistics (e.g., mean, median, standard deviation, etc.) will be calculated as appropriate. The Principal Investigator for TK data analysis will be Ann-Muriel Steff, PharmD, Ph.D.

Principal Investigator: PharmD, Ph.D.
Senior Research Scientist, Pharmacokinetics
Charles River Preclinical and Clinical Services
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA
Tel: +1.514.630.8200, ext. 8368
Fax: +1.514.630.8230
E-mail:

22. METHOD OF SACRIFICE

Mice will be sacrificed by carbon dioxide asphyxiation.

22.1. Scheduled Sacrifice

Mice will be sacrificed according to the blood sample collection timepoints and discarded without further evaluation.

22.2. Mice Found Dead or Unscheduled Sacrifice

Mice that die or are sacrificed before scheduled termination will be examined for the cause of death or condition as soon as possible after the observation is made. Whenever possible, blood samples will be obtained from mice sacrificed before scheduled termination. Samples will be collected, processed and shipped as described in section 21.5.

23. PROPOSED STATISTICAL METHODS

Averages and percentages will be calculated. Additional procedures and/or analyses may be performed, if appropriate.

24. DATA ACQUISITION, VERIFICATION AND STORAGE

Data generated during the course of this study will be recorded either by hand or using the *Argus Automated Data Collection and Management System,* the *Vivarium Temperature and Relative Humidity Monitoring System* and/or chart recorders. All data will be tabulated, summarized and/or statistically analyzed using the *Argus Automated Data Collection and Management System*, the *Vivarium Temperature and Relative Humidity Monitoring System, Microsoft® Excel* (part of Microsoft® Office 97/2000/2003/XP), Quattro Pro 8 and/or *The SAS System* (version 6.12).

Records will be reviewed by the Study Director and/or appropriate management personnel within 21 days after generation. All original records will be stored in the archives at the Testing Facility. All raw data will be bound and indexed. The archived raw data will be scanned and retained on CD-ROM in an Adobe® Acrobat PDF file. A copy of all raw data will be supplied to the Sponsor upon request. Preserved tissues will be stored at the Testing Facility for ten years after mailing of the draft final report, after which time the Sponsor will be contacted to determine the disposition of these materials.

25. RECORDS TO BE MAINTAINED

Protocol and Amendments.
Test Substance, Vehicle and/or Reagent Receipt, Preparation and Use.
Animal Acquisition.
Randomization Schedules.
Supportive Care (if prescribed by Staff Veterinarian).
General Comments.
Clinical Observations and/or General Appearance.
Blood Sample Collection, Processing and Shipment.
Body Weights.
Photographs (if required).
Study Maintenance (room and environmental records).
Feed and Water Analyses.
Packing and/or Shipment Lists.
Necropsy (if required).

26. KEY PERSONNEL

Director of Research and Study Director: , Ph.D., DABT, Fellow ATS
Director of Reproductive and Neurobehavioral Toxicology: , Ph.D.
Director of Operations: , B.S.
Associate Director of Regulatory Compliance: , M.S.
Senior Manager of Study Management: , B.S., RQAP-GLP, ALAT
Senior Staff Veterinarian: V.M.D., Division Veterinarian
Chair, Institutional Animal Care and Use Committee: , B.S., LAT
Consultant, Veterinary Pathology: D.V.M., Ph.D., Diplomate, ACVP

27. FINAL REPORT

The Study Director will provide periodic updates of study progress to the Sponsor. Draft summary tables of unaudited computer-recorded data may accompany these updates. Statistical analyses will not be performed on these interim data.

A comprehensive draft final report will be prepared on completion of the study and will be finalized following consultation with the Sponsor. The report will include the following:

Summary and Conclusion.
Experimental Design and Method.
Evaluation of Test Results.
Appendices: Figures, Summary and Individual Tables Summarizing the Above Data, Protocol and Associated Amendments and Deviations, Study Director's GLP Compliance Statement, Reports of Supporting Data (if appropriate) and QAU Statement.

28. ANIMAL WELFARE

Animal care and use will be in accordance with the Animal Welfare Act regulations (9 CFR, Parts 1, 2 and 3), the conditions specified in the *Guide for the Care and Use of Laboratory Animals*[4], the relevant SOPs of the Testing Facility, and the protocol. Anticipated or suspected clinical signs and supportive care agreed upon by the Study Director, veterinary staff and Sponsor should these clinical signs be observed are documented in the IACUC proposal for this study.

Adverse observations will be promptly reported to the Study Director and veterinary staff. The veterinarian may make recommendations regarding care of the animal(s) in addition to those already agreed upon and/or alteration of study procedures to ensure the well-being of the animal(s) should unanticipated responses or circumstances occur. All recommendations shall be discussed with the Study Director and the recommendations and subsequent actions properly documented in the study record. Supportive care of the animal(s) may occur without notification of the Sponsor when such supportive care, as determined by the Study Director, does not adversely affect the study objectives.

If the condition of the animal(s) warrants therapeutic intervention or alterations in study procedures above the previously-agreed-upon conditions, the Sponsor will be contacted, whenever possible, to discuss appropriate action. If the condition of the animal(s) is such that immediate measures must be taken to relieve pain and/or distress, the attending veterinarian will attempt to consult the Study Director prior to initiating medical action, but the veterinarian has the authority to act immediately at his/her discretion to address the condition under these circumstances. The Sponsor will be informed by the Study Director of any such event as soon as possible.

29. INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE STATEMENT

The procedures described in this protocol have been reviewed by the Testing Facility's Institutional Animal Care and Use Committee. All procedures described in this protocol that involve study animals will be conducted in a manner to avoid or minimize discomfort, distress or pain to the animals.

The signature of the Sponsor's representative below is assurance that the study is not an unnecessary duplication of previous work. Documentation for the necessity of this study may be obtained from the Sponsor. No alternative procedures were available to meet the stated purposes of the study.

30. REFERENCES

(1) Federal Insecticide, Fungicide and Rodenticide Act/Toxic Substances Control Act (FIFRA/TSCA); Good laboratory practice standards; Final Rule 40 C.F.R Part 160/792; August 17, 1989. U.S. Environmental Protection Agency.

(2) Good laboratory practice standards for toxicological studies on agricultural chemicals. 59-Nousan-No.3850; August 10, 1984. Repealed as 1 October, 1999. Notification 11-Nousan-No.6283. Japan: Ministry of Agriculture, Forestry and Fisheries, Japan (MAFF).

(3) OECD Principles of good laboratory practices, [C(97)186/Final] (1998); Environmental Health and Safety Division. OECD Environment Directorate.

(4) Institute of Laboratory Animal Resources Commission on Life Sciences and the National Research Council. *Guide for the care and use of laboratory animals.* Washington (D.C.): National Academy Press; 1996.

(5) Dar, K., Grey, B., Zehr, B., Wood, C., Butenhoff, J., Chang, S., Ehresman, D., Tan, Y. & Lau, C. 2008, Effects of Perfluorobutyrate Exposure during Pregnancy in the Mouse, Toxicological Sciences 105(1), 173-181 (2008).

31. PROTOCOL APPROVAL

31.1. For The Testing Facility

---------- 20 May 09
Date

Director of Reproductive and
Neurobehavioral Toxicology
Testing Facility Management

20 May 09
Date

Director of Research
Study Director

31.2. For The Sponsor[a]

22 May 09
Date

Toxicologist
Study Monitor

a. Date of Sponsor Approval: 20 May 2009

Original could not be located. This copy will serve as the original. 11 MAR 2010 AD

ATTACHMENT 1 -

PROPOSED STUDY SCHEDULE

PROPOSED SCHEDULE[a]

19 MAY 2009	Animals Arrive
19 MAY 2009	Experimental Start Date - OECD
27 MAY 2009	Experimental Start Date - EPA
19 MAY 2009 - 26 MAY 2009	Acclimation Period
27 MAY 2009	Dosage Administration
27 MAY 2009 - 28 MAY 2009	Blood Sample Collection and Scheduled Sacrifice
01 JUN 2009	Shipment of Blood Samples.
09 JUL 2009	Submission Date of Audited Draft Report.
06 JUL 2009	Experimental Completion / Termination Date.

---

a. The study initiation date is the date the Study Director signs the protocol.

ATTACHMENT 2 -

MATERIAL SAFETY DATA SHEET

DAIKIN

Safety Data Sheet
according to 1907/2006/EC, Article 31

Printing date 28.11.2007 *Revision: 30.11.2005*

1 Identification of the substance/preparation and of the company/undertaking

· **Product details**

· **Trade name: PFH Ammonium Salt (C-1500N)**

· **Application of the substance / the preparation** Emulsifier

· **Manufacturer/Supplier:**
DAIKIN INDUSTRIES, LTD.CHEMICAL DIVISION:
Umeda Center Bldg., 4-12, Nakazaki-Nishi2-chome, Kita-Ku, Osaka, JAPAN
Phone: (+81) 6-6373-4349 Fax: (+81) 6-6373-4389

· **Further information obtainable from:** http://www.daikin.co.jp/chm/en/index.html
· **Information in case of emergency:** +81-6-6349-7521, +49-211-179 225, 1-256-306-5000

2 Hazards identification

· **Hazard description:**

Xi Irritant

· **Information concerning particular hazards for human and environment:**
The product has to be labelled due to the calculation procedure of the "General Classification guideline for preparations of the EU" in the latest valid version.
R 41 Risk of serious damage to eyes.
· **Classification system:**
The classification is according to the latest editions of the EU-lists, and extended by company and literature data.

3 Composition/information on ingredients

· **Chemical characterization**
· **Description:** liquid solution. water 50%

· **Dangerous components:**			
21615-47-4	Ammonium Perfluorohexanoate	Xi; R 41	50.0%

· **Additional information:** For the wording of the listed risk phrases refer to section 16.

4 First-aid measures

· **After inhalation:** Supply fresh air; consult doctor in case of complaints.
· **After skin contact:** Generally the product does not irritate the skin.
· **After eye contact:** Rinse opened eye for several minutes under running water. Then consult a doctor.
· **After swallowing:** If symptoms persist consult doctor.

5 Fire-fighting measures

· **Suitable extinguishing agents:**
CO2, powder or water spray. Fight larger fires with water spray or alcohol resistant foam.
· **Special hazards caused by the substance, its products of combustion or resulting gases:**
Hydrogen fluoride (HF)
Formation of toxic gases is possible during heating or in case of fire.
· **Protective equipment:** Wear fully protective suit.

GB

(Contd. on page 2)

Safety Data Sheet
according to 1907/2006/EC, Article 31

Printing date 28.11.2007 Revision: 30.11.2005

Trade name: PFH Ammonium Salt (C-1500N)

(Contd. of page 1)

6 Accidental release measures

· **Person-related safety precautions:**
There is no acute toxic risk known to be associated with this substance. Use self-contained respiratory protective device and non-permeable gloves are recommended against inhalation and transdermal uptake (see attached ppt file)
· **Measures for environmental protection:**
Dilute with plenty of water.
Do not allow to enter sewers/ surface or ground water.
· **Measures for cleaning/collecting:**
Absorb with liquid-binding material (sand, diatomite, acid binders, universal binders, sawdust).
Ensure adequate ventilation.

7 Handling and storage

· **Handling:**
· **Information for safe handling:**
Ensure good ventilation/exhaustion at the workplace.
Prevent formation of aerosols.
See attached ppt file.
· **Information about fire - and explosion protection:** No special measures required.

· **Storage:**
· **Requirements to be met by storerooms and receptacles:** No special requirements.
· **Information about storage in one common storage facility:** Not required.
· **Further information about storage conditions:** None.

8 Exposure controls/personal protection

· **Additional information about design of technical facilities:** No further data; see item 7.

· **Ingredients with limit values that require monitoring at the workplace:**
The product does not contain any relevant quantities of materials with critical values that have to be monitored at the workplace.
· **Additional information:** The lists valid during the making were used as basis.

· **Personal protective equipment:**
· **General protective and hygienic measures:**
Immediately remove all soiled and contaminated clothing
Wash hands before breaks and at the end of work.
Avoid contact with the eyes.
· **Respiratory protection:**
In case of brief exposure or low pollution use respiratory filter device. In case of intensive or longer exposure use self-contained respiratory protective device.
· **Protection of hands:**
The glove material has to be impermeable and resistant to the product/ the substance/ the preparation.
Due to missing tests no recommendation to the glove material can be given for the product/ the preparation/ the chemical mixture.
Selection of the glove material on consideration of the penetration times, rates of diffusion and the degradation
· **Material of gloves**
Double glove, supported nitrile or neoprene over latex under-glove, recommended for extended use. Gloves should be discarded at end of use if soiled.
· **Penetration time of glove material**
The exact break trough time has to be found out by the manufacturer of the protective gloves and has to be observed.

(Contd. on page 3)

Safety Data Sheet
according to 1907/2006/EC, Article 31

Printing date 28.11.2007 Revision: 30.11.2005

Trade name: PFH Ammonium Salt (C-1500N)

(Contd. of page 2)

· **Eye protection:**

Tightly sealed goggles

· **Body protection:** Protective work clothing

9 Physical and chemical properties

· **General Information**	
Form:	Solution
Colour:	Colourless
Odour:	Aromatic
· **Change in condition**	
Melting point/Melting range:	Undetermined.
Boiling point/Boiling range:	100°C
· **Flash point:**	Not applicable.
· **Self-igniting:**	Product is not selfigniting.
· **Danger of explosion:**	Product does not present an explosion hazard.
· **Vapour pressure at 20°C:**	23.0 hPa
· **Density:**	Not determined.
· **Solubility in / Miscibility with water:**	Fully miscible.
· **pH-value at 20°C:**	7.0
· **Solvent content:**	
Organic solvents:	0.0 %
Water:	50.0 %
· **Solids content:**	50.0 %

10 Stability and reactivity

· **Thermal decomposition / conditions to be avoided:** No decomposition if used according to specifications.
· **Dangerous reactions** No dangerous reactions known.
· **Dangerous decomposition products:**
Hydrogen fluoride
Fluorophosgene on contact with naked flame or red hot objects.

11 Toxicological information

· **Acute toxicity:**

· **LD/LC50 values relevant for classification:**		
21615-47-4 Ammonium Perfluorohexanoate		
Oral	LD50	>2000 mg/kg (rat)
Dermal	LD50	>2000 mg/kg (rat)

· **Primary irritant effect:**
· **on the skin:** No irritant effect.
· **on the eye:** Strong irritant with the danger of severe eye injury.

(Contd. on page 4)

Safety Data Sheet
according to 1907/2006/EC, Article 31

Printing date 28.11.2007 Revision: 30.11.2005

Trade name: PFH Ammonium Salt (C-1500N)

(Contd. of page 3)

· **Sensitization:** No sensitizing effects known.
· **Additional toxicological information:**
The product shows the following dangers according to the calculation method of the General EU Classification Guidelines for Preparations as issued in the latest version:
Irritant
· **Toxicokinetics, metabolism and distribution**
ADME
Rat : Male : T1/2 = 1.0 hr, Female : T1/2 = 0.42 hr
Monkey : Male : T1/2 = 5.3 hr, Female : T1/2 = 2.4 hr
· **Repeated dose toxicity**
90-day oral toxicity in rodents
Male NOEL = 10 mg/kg/day (body weight loss at >50 mg/kg, lower Cholesterol and Ca)
Female NOEL = 50 mg/kg/day (lower globulin at 200 mg/kg)
· **CMR effects (carcinogenity, mutagenicity and toxicity for reproduction)**
Combined repeated dose toxicity with the reproduction/development toxicity screening test
Reproductive(OECD TG 422)
Male & Female NOAEL = 300,450 mg/kg/day (F1:no reproductive changes)

12 Ecological information

· **Information about elimination (persistence and degradability):**
· **Other information:** The product is difficultly biodegradable.
· **Ecotoxical effects:**
· **Acquatic toxicity:**
Acute toxicity to Daphnia magna
24 hr EC50 = >100 mg/L
48 hr EC50 = >100 mg/L
NOEC = >100 mg/L

Acute toxicity to Fish
96 hr LC50 = >100 mg/L
NOEC = >100 mg/L "

Algal inhibition test
72 hr EbC50 = 90 mg/L
0-72 hr ErC50 = 86 mg/L
NOEC = 50 mg/L
· **General notes:**
Water hazard class 1 (German Regulation) (Self-assessment): slightly hazardous for water
Do not allow undiluted product or large quantities of it to reach ground water, water course or sewage system.

13 Disposal considerations

· **Product:**
· **Recommendation**
Must not be disposed together with household garbage. Do not allow product to reach sewage system.

· **Uncleaned packaging:**
· **Recommendation:** Disposal must be made according to official regulations.
· **Recommended cleansing agents:** Water, if necessary together with cleansing agents.

GB

(Contd. on page 5)

Safety Data Sheet
according to 1907/2006/EC, Article 31

Printing date 28.11.2007 *Revision: 30.11.2005*

Trade name: PFH Ammonium Salt (C-1500N)

(Contd. of page 4)

14 Transport information

· **Land transport ADR/RID (cross-border)**
· **ADR/RID class:** -

· **Maritime transport IMDG:**
· **IMDG Class:** -
· **Marine pollutant:** No

· **Air transport ICAO-TI and IATA-DGR:**
· **ICAO/IATA Class:** -

15 Regulatory information

· **Labelling according to EU guidelines:**
Observe the general safety regulations when handling chemicals.
The product has been classified and marked in accordance with EU Directives / Ordinance on Hazardous Materials.

· **Code letter and hazard designation of product:**
Xi Irritant

· **Risk phrases:**
41 Risk of serious damage to eyes.

· **Safety phrases:**
23 Do not breathe gas/fumes/vapour/spray (appropriate wording to be specified by the manufacturer).
26 In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
39 Wear eye/face protection.
60 This material and its container must be disposed of as hazardous waste.

· **National regulations:**

· **Waterhazard class:** Water hazard class 1 (Self-assessment): slightly hazardous for water.

16 Other information

This information is based on our present knowledge. However, this shall not constitute a guarantee for any specific product features and shall not establish a legally valid contractual relationship.

· **Relevant R-phrases**
41 Risk of serious damage to eyes.

· **Department issuing MSDS:** Toxicology and Product Regulatory
· **Contact:** www.daikin.co.jp/chm/

GB

ATTACHMENT 3 -

TEST SUBSTANCE PREPARATION PROCEDURE

ATTACHMENT 3

TEST SUBSTANCE PREPARATION PROCEDURES

Test Substance: PFH Ammonium Salt (Ammonium salt of Perfluorinated Hexanoic Acid); supplied as a 50% aqueous solution

Vehicle: R.O. deionized water

A. Purpose:

The purpose of this procedure is to provide a method for the preparation of the dosage solutions of the test substance for oral (gavage) administration to mice on Study No. UZS00009.

B. General Information:

1. All solution containers will be labeled and color-coded. Each label will specify the study number, vehicle or test substance identification, batch number, concentration, dosage level, dosage group, preparation date, expiration date, and storage conditions, as applicable.

2. Formulations (solutions) of the test substance will be prepared at least once at the Testing Facility by direct dilution of the Sponsor-supplied stock test substance solution with the vehicle and used within 48 hours of formulation.

3. Formulations (solutions) will be administered at a final dosage volume of 5 mL/kg.

4. Safety:

X Double nitrile gloves, uniform/lab coat, goggles or safety glasses with side shields
X Dust mist/HEPA-filtered Mask
___ Half-Face Respirator
___ Full-Face Respirator/Positive Pressure Hood
X Tyvek® Sleeves

5. The test substance will be considered 95% by weight of PFH acid for the purpose of dosage calculations.

6. Sampling requirements: Cited in protocol
7. Storage: Cited in protocol

C. Preparation of the Test Substance Dosage Solutions:

1. Add the required amount of vehicle to an appropriately sized and labeled container (See TA/S DILUTION CALCULATION SHEET).

2. Add the required amount of the Sponsor-supplied stock test substance solution to the container (See TA/S DILUTION CALCULATION SHEET).

3. Add a magnetic stir bar to the container. Place the container on a magnetic stir plate and mix continuously for at least 24 hours prior to sampling and/or aliquotting.

4. Aliquot the formulation into an appropriate number of appropriately sized and labeled containers. Aliquots will be stored at room temperature.

5. On the day prior to dosage administration, remove the required number of aliquots from storage. Add a magnetic stir bar to the container. Place the container on a magnetic stir plate and stir continuously **for at least 24 hours** prior to dosage administration. Continue to mix the formulation during dosage administration. Any formulation remaining after being used for dosage administration will be discarded at the Testing Facility.

6. Repeat steps C1 through C5 for each concentration.

Version: UZS00009(14.MAY.2009) # of pages: 2

Prepared By: Date: 20MAY2009

Approved By: Date: 20 MAY 09

charles river

Amendment 1

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

Charles River Laboratories Study No. UZS00009

Item No. 1.

Protocol Section: ATTACHMENT 3 - TEST SUBSTANCE PREPARATION PROCEDURE (Page 32 of the protocol)

Change: The test article preparation procedure attached to the protocol [UZS00009(14.MAY.2009)] is being replaced with the attached test article preparation procedure [UZS00009 (22.MAY.2009)].

Justification: The new preparation procedure [UZS00009 (22.MAY.2009)] replaces the previous [UZS00009(14.MAY.2009)] preparation procedure and corrects the procedure from a weight/volume (mg/ml) procedure to a weight/weight (mg/g) procedure.

TESTING FACILITY SIGNATURES

7 Jul
Date

Director of Research.
Study Director

905 Sheehy Drive, Bldg. A, Horsham, PA 19044 • 215.443.8710 • FAX 215.443.8587

ATTACHMENT 3

TEST SUBSTANCE PREPARATION PROCEDURES

Test Substance: PFH Ammonium Salt (Ammonium salt of Perfluorinated Hexanoic Acid); supplied as a 50% aqueous solution

Vehicle: R.O. deionized water

A. Purpose:

The purpose of this procedure is to provide a method for the preparation of the dosage solutions of the test substance for oral (gavage) administration to mice on Study No. UZS00009.

B. General Information:

1. All solution containers will be labeled and color-coded. Each label will specify the study number, vehicle or test substance identification, batch number, concentration, dosage level, dosage group, preparation date, expiration date, and storage conditions, as applicable.

2. Formulations (solutions) of the test substance will be prepared at least once at the Testing Facility by direct dilution of the Sponsor-supplied stock test substance solution with the vehicle and used within 48 hours of formulation.

3. Formulations (solutions) will be administered at a final dosage volume of 5 mL/kg.

4. Safety:

X Double nitrile gloves, uniform/lab coat, goggles or safety glasses with side shields
X Dust mist/HEPA-filtered Mask
___ Half-Face Respirator
___ Full-Face Respirator/Positive Pressure Hood
X Tyvek® Sleeves

5. The test substance will be considered 95% by weight of PFH acid for the purpose of dosage calculations.

6. Sampling requirements: Cited in protocol

7. Storage: Cited in protocol

C. Preparation of the Test Substance Dosage Solutions:

1. Add the required amount of the Sponsor-supplied stock test substance solution (in grams) to a volumetric flask (See TA/S DILUTION CALCULATION SHEET).

2. Q.S. to the desired volume with vehicle in the volumetric flask. (See TA/S DILUTION CALCULATION SHEET).

3. Add a magnetic stir bar to the flask. Place the flask on a magnetic stir plate and mix continuously for at least 24 hours prior to and during sampling and/or aliquotting.

4. Aliquot the formulation into an appropriate sized and labeled container. Aliquots will be stored at room temperature and stirred continuously. Continue to mix the formulation during dosage administration. Any formulation remaining after being used for dosage administration will be discarded at the Testing Facility.

5. Repeat steps C1 through C5 for each concentration.

Version: UZS00009(22.MAY.2009) # of pages: 2

Prepared By: Date: 22 may 2009

Approved By: Date: 22 may 09

Amendment 2

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

Charles River Laboratories Study No. UZS00009

Item No. 1.

Protocol Section: Regulatory Compliance (Page 6 of the protocol)

Change: A process audit, rather than a critical phase inspection, will be performed for analysis of the bulk test substance.

Justification: This change is being made to clarify the type of auditing that will be performed for the bulk test substance analysis.

Item No. 2.

Protocol Section: Bulk Test Substance Stability (Page 9 of the protocol)

Change: Stability will be assessed by normalization purity by HPLC and the value compared to the purity identified on the Certificate of Analysis. A report will be generated for this phase of the study and provided to the Study Director for inclusion in the final report.

Justification: This change is being made to describe how analysis of the bulk test substance will be performed and to clarify that the phase report will be provided and included in the final report for the study.

905 Sheehy Drive, Bldg. A, Horsham, PA 19044 • 215.443.8710 • FAX 215.443.8587

Item No. 3.

Protocol Section: Analysis of Prepared Formulations (Page 9 of the protocol)

Change: Formulation analysis will be performed using Good Laboratory Practice (GLP)-validated HPLC method number (performed as Charles River Laboratories Preclinical Services Montreal Study number 211052). The Test Site Reference number for the work in this current study is 211053. A report will be generated for this phase of the study and provided to the Study Director for inclusion in the final report.

Justification: This change is being made to identify the type of method used for formulation analysis and the associated Test Site reference numbers, and to clarify that the phase report will be provided and included in the final report for the study.

Item No. 4.

Protocol Section: Data Acquisition, Verification and Storage (Page 17 of the protocol)

Change: Empower (Waters Corporation) will be used for formulation sample analysis.

Justification: This change is being made to identify the system used for formulation sample analysis.

TESTING FACILITY SIGNATURES

12-Nov-09
Date

Director of Research.
Study Director

Amendment 3

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

Charles River Laboratories Study No. UZS00009

Item No. 1.

Protocol Section: Pharmacokinetic Analysis Scientist (Page 5 of the protocol and citations throughout)

Change: The Principal Investigator for Pharmacokinetic Analysis will be Thanh Do.

Principal Investigator:
Research Scientist, Bioanalysis
Charles River Preclinical and Clinical Services
22022 Transcanadienne, Senneville
Quebec H9X 3R3
CANADA
Tel: +1.514.630.8200 ext. 2224
Fax:

Justification: The Principal Investigator for Pharmacokinetic Analysis was to be added by amendment.

Item No. 2.

Protocol Section: Shipping Instructions (Page 15 of the protocol)

Change: The text in bold is being added to this section.

Principal Investigator:

d Clinical Services
22022 Transcanadienne Senneville
Montreal, Quebec H9X 3R3
CANADA
Custom Clearance: H. Kennedy Inc
Tel: +1.514.630.8200 ext 8974
Fax: +1.514.630.8200
E-mail

Justification: The Principal Investigator was to be added by amendment.

Item No. 3.

Protocol Section: Pharmacokinetic Sample Analysis (Page 15 of the protocol)

Change: The samples will be analyzed at PCS-MTL using a validated LC-MS/MS method (PCS-MTL Study No. 141658). The bioanalytical method will be validated to meet the minimum requirements of the appropriate PCS-MTL Standards Operating Procedures. Remaining unused study samples will be retained at PCS-MTL for approximately 1 year after dispatch of the final report or until authorized to discard by the Study Director. Incurred sample reanalysis (ISR) will be performed for this study as per the appropriate PCS-MTL SOP.

Justification: This change was to be added by amendment.

Item No. 4.

Protocol Section: Data Acquisition, Verification and Storage (Page 17 of the protocol)

The following text is being added to this section.

Change: Data collection for serum concentration analysis using LC-MS/MS will be performed using Analyst from MDS Sciex. Statistical analysis, including regression analysis, and descriptive statistics such as arithmetic means and standard deviations, accuracy and precision will be performed using Watson Laboratory Information Management system (LIMS) and Microsoft Excel. Tables will be prepared from retrospective manual entry on computer (Microsoft Word).

All raw data and documents generated at PCS-MTL during this study and the final report will be transferred to the scientific archives of PCS-MTL for a period of approximately 1 year from finalization. Storage details following the 1 year archive period will be documented in the raw data.

Justification: This addition identifies statistical analysis and archival information for data generated at PCS-MTL.

TESTING FACILITY SIGNATURES

TVL-01
Date

Director of Research
Study Director

Protocol Amendment No. 4

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

Testing Facility Study No. UZS00009

1. Amendment 3, Item No. 3

The samples will be analyzed at PCS-MTL using a validated LC-MS/MS method (PCS-MTL Study No. **141837**). The bioanalytical method will be validated to meet the minimum requirements of the appropriate PCS-MTL Standards Operating Procedures. Remaining unused study samples will be retained at PCS-MTL for approximately 1 year after dispatch of the final report or until authorized to discard by the Study Director. Incurred sample reanalysis (ISR) will be performed for this study as per the appropriate PCS-MTL SOP.

Justification:

The initial analytical method had inadequate analytical range for the analysis of the mouse serum samples. Therefore, a new method was validated with a higher analytical range. The validation number is being updated to reference the new method which will be used to analyze the mouse serum samples.

Protocol Amendment No. 4

Amendment Approval:

2 Oct 09
Date

Executive Director, Site Operations and Toxicology
Study Director

Protocol Amendment No. 5

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid) in Mice

Testing Facility Study No. UZS00009
All procedures will be conducted as outlined in the original protocol except for the following changes regarding the study extension.

1. Section 3. Purpose

The purpose of this study extension is to analyze the test substance levels in the livers of Crl:CD1(ICR) female mice.

Justification:

The purpose is being added to identify the study extension. The study is being extended in order to attain the appropriate range for validation of the assay in the liver. Additionally, this information may be used in determining dosage levels for the segment II/III toxicity study in mice.

2. Section 11. Regulatory Compliance

The extended portion of this study, designed for validation of the assay in the liver will be conducted using good scientific practices and according to the Standard Operating Procedures (SOPs) of the Testing Facility. The Testing Facility Quality Assurance Unit (QAU) will not audit this portion of the study.

Justification:

The study extension will be conducted non-GLP.

3. Section 17.2. Number

Population selected for study extension: 6 female mice (2 per dosage group).

Justification:

This change identifies the number of mice assigned to the extension portion of the study.

4. Section 17.4. Body Weight and Age

Female mice will be approximately 180 days of age at transfer, at which time they will be expected to have body weights of 30 g to 50 g each. Actual body weights and age will be recorded the day after transfer and documented in the raw data.

Justification:

This change identifies the body weight and age of the test system.

5. Section 17.5. Source

The mice will be transferred to study from the Testing Facility general population.

Justification:

This change identifies the source of the test system.

6. Section 19. Randomization

Upon transfer, female mice will be assigned to individual housing on the basis of computer-generated random units. Female mice will be assigned to study groups using a second computer-generated (weight-ordered) randomization procedure on the day before dosing.

Justification:

This change identifies the randomization procedures for the extension study.

7. Section 21.2 Clinical Observations and/or General Appearance

Dosage Period: Prior to dosage administration. Postdosage observations will be recorded prior to sacrifice.

Justification:

This change identifies clinical observations during the dosage period for the study extension.

8. Section 21.5 Pharmacokinetic Sample Collection

Two hours (± 10 minutes) after dosage administration, livers will be excised and retained frozen ($\leq -70^{o}C$) from all mice after sacrifice. The time of each liver collection will be documented in the raw data.

Justification:

This change identifies the pharmacokinetic sample collection.

9. Section 21.6 Pharmacokinetic Sample Analysis

Liver samples will be analyzed using a non-validated LC-MS/MS for determination of suitable analytical range.

Justification:

This change identifies the method of sample analysis.

10. Section 22.1. Scheduled Sacrifice

Mice will be sacrificed via carbon dioxide asphyxiation 2 hours ± 10 minutes after dosage administration. Livers will be excised and stored frozen (≤ -70°C) until shipment for analysis. Carcasses will be discarded without further evaluation.

The recipient will be notified in advance of sample shipment. Sample will be shipped (on dry ice) to:

Principal Investigator:
Research Scientist, Bioanalysis
Charles River Preclinical and Clinical Services
22022 Transcanadienne, Senneville
Quebec H9X 3R3
CANADA
Tel: +1.514.630.8200 ext. 2224
Fax: +1.514.630.8230
E-mail:

Justification:

This change identifies the endpoint of the study extension.

11. Section 22.2. Mice Found Dead or Unscheduled Sacrifice

Mice that die or are sacrificed before scheduled termination will be examined for the cause of death or condition as soon as possible after the observation is made. The liver will be excised and retained frozen (≤ -70°C) until shipment for analysis. Carcasses will be discarded without further evaluation.

Justification:

This change identifies the procedures for mice deceased before scheduled termination.

12. Attachment 1 - Proposed Study Schedule

09 NOV 2009	Animals Transferred
11 NOV 2009	Dosage Administration
11 NOV 2009	Scheduled Sacrifice (Approximately 2 hours ± 10 minutes postdosage)
16 NOV 2009	Shipment of Tissues.

Justification:

This change identifies the proposed study schedule.

Amendment Approval:

Date: 5-NOV-09

Executive Director, Site Operations and Toxicology
Study Director

APPENDIX 2 - CERTIFICATE OF ANALYSIS

Certificate of Analysis

Daikin Industries,LTD.

Name of Sample	*PFH Ammonium Salt（C-1500N）*
Lot.	*7005*
Date of Analysis	*May 14, 2009*
Purify	*47.4% (Effective component in Water)*
	**50.8*0.934%=47.4%*

COMPOSITION

identity		*Conc.*
#1	*Ammonium Perfluorohexanoate CAS RN. 21615-47-4*	*93.4%*
#2	*Unknown*	*6.6%*
	Total	*100%*

Analysis system (HPLC)

Equipment	*: Waters Alliance2695*
Detector	*: Waters 2487UV*
Detection wavelength	*: 210nm*

Analysis condition

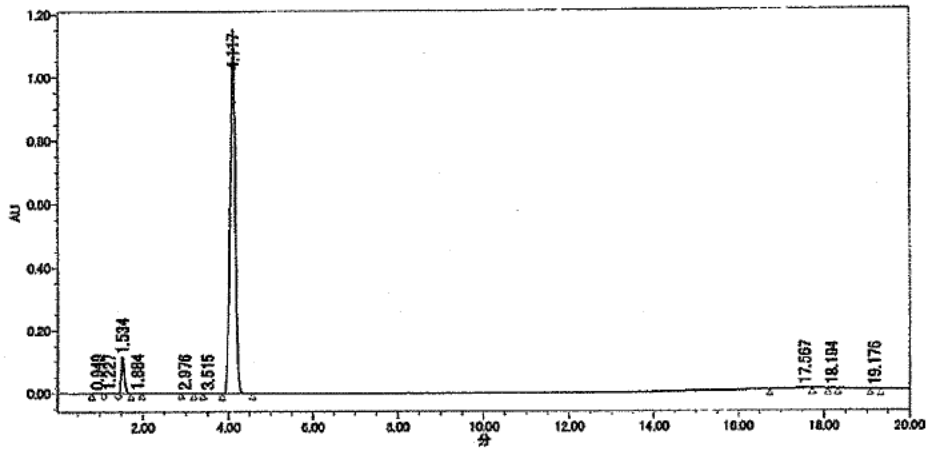
Column	*: TOSOH TSKGel ODS120T 4.6mm×150mm*
Temp.	*: 40 ℃*
Mobile phase	*: A=acetonitrile 、B=Solution of 0.6% perchloric acid in water*
Gradient	*: A:B=50:50(mass%) (0-10min.) →90:10(mass%) (15-20min.)*
Injection volume	*: 20μL*
Injection Concentration	*: 1%（dilute 50times with water)*

Chemical R&D Center
Unidyne Group
Senior Researcher

SIGNATURE
DATE : May 18, 2009

Analysis

サンプル名:	C1500N	分析担当者:	System
サンプルの種類:	未知試料	分析日:	2009/05/14 11:49:44
バイアル:	82	取り込みメソッドセット:	090514S
注入#:	1	解析日:	2009/05/14 13:55:17
注入量:	20.00 μl	解析メソッド:	C1500N
分析時間:	20.00 分	チャンネル名:	2487チャンネル 1
サンプルセット名:		解析チャンネルの説明:	



	成分名	Retention Time(min.)	Area (μVsec.)	Area (%)	Height (μV)		
1		0.949	17634	0.18	2554		
2		1.227	20551	0.20	1927		
3		1.534	574660	5.71	116134		
4		1.884	5543	0.06	710		
5		2.976	2424	0.02	414		
6		3.515	4940	0.05	361		
7		4.117	9390042	93.38	1144218		
8		17.567	29475	0.29	984		
9		18.194	6956	0.07	1098		
10		19.176	3881	0.04	592		

1/1 ページ

APPENDIX 3 - ANALYTICAL REPORT

FINAL REPORT

Test Site Ref. No. 211053
Testing Facility Study No. UZS00009

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium Salt of Perfluorinated Hexanoic Acid) in Mice

TEST SITE:
Charles River Laboratories Preclinical Services Montreal
22022 Transcanadienne
Senneville, Quebec
Canada H9X 3R3

TESTING FACILITY:
Charles River Laboratories Preclinical Services
905 Sheehy Drive, Building A
Horsham, PA 19044
USA

SPONSOR:
Daikin Industries, Ltd.
Chemical Division
Umeda Center Building
4-12 Nakazaki-Nishi, 2-chrome
Kita-ku, Osaka 530-8323
Japan

31 Mar 2010

Page 1 of 29

TABLE OF CONTENTS

LIST OF TABLES 3
LIST OF FIGURES 4
LIST OF APPENDICES 5
1. COMPLIANCE STATEMENT 6
2. QUALITY ASSURANCE STATEMENT 7
3. SUMMARY 8
4. INTRODUCTION 9
5. REFERENCE STANDARD AND VEHICLE 9
5.1. Reference Standard 9
5.2. Vehicle 9
6. EXPERIMENTAL PROCEDURES 10
6.1. Standard Stock Solutions 10
6.2. Standard Solutions 10
6.3. Spiked Samples 10
6.4. Study Samples 10
6.5. Bulk Test Substance Stability 11
6.6. Analysis 11
6.7. System Suitability 12
6.8. Data Collection and Statistical Methods 12
6.9. Quality Assurance 13
6.10. Archives 13
7. PROTOCOL DEVIATIONS 13
8. RESULTS AND DISCUSSION 13
8.1. System Suitability 13
8.2. Study Samples 14
8.3. Bulk Test Substance Stability 14
9. CONCLUSION 14
10. REPORT REVIEW AND APPROVAL SIGNATURE 15

LIST OF TABLES

Table 1 Study Samples - Concentration and Homogeneity 16

Table 2 Study Samples - Stability Assessment Following 10 Days Storage at Room Temperature 17

Table 3 Bulk Substance Stability (48 Days Storage at Room Temperature)..... 18

LIST OF FIGURES

Figure 1 Representative Standard Chromatogram (Nominal Concentration: 190 µg/mL) 19

Figure 2 Representative Blank Vehicle 20

Figure 3 Representative Sample Chromatogram (Group 1, Top, Sampling Date: 26 May 2009, Nominal Concentration: 7 mg/mL; Nominal Injected Concentration: 105 µg/mL) 21

Figure 4 Representative Bulk Substance Sample (Full Scale) 22

Figure 5 Representative Bulk Substance Sample (Auto-scaled) 23

Figure 6 Representative Blank Sample (Auto-scaled) 24

LIST OF APPENDICES

Appendix 1 Certificate of Analysis 25

Appendix 2 Principal Investigator Acknowledgment Form 28

1. COMPLIANCE STATEMENT

This portion of the study, conducted at Charles River Laboratories Preclinical Services Montreal (PCS-MTL), 22022 Transcanadienne, Senneville, Quebec, Canada, H9X 3R3, complied with the Good Laboratory Practice (GLP) Regulations of the United States Food and Drug Administration (21 CFR Part 58), the U.S. Environmental Protection Agency and the principles of the Organization for Economic Co-operation and Development (OECD). However, the work was not conducted as per the Ministry of Agriculture Forestry and Fisheries, due to the fact the test site is not accredited by this agency.

_____ 24 Mar 2010

Date

Principal Investigator
Research Scientist, Analytical Chemistry
Laboratory Sciences
Charles River Laboratories

2. QUALITY ASSURANCE STATEMENT

In compliance with the Good Laboratory Practice Regulations, Reference No. 211053 has been audited. The data presented in the final report accurately represent the data collected during the conduct of the study.

Phase or Segment Audited	Date of Inspection	Dates of Reports to Test Site Management and Principal Investigator	Dates of Reports to Testing Facility Management/ Study Director & Lead QA
Analytical Chemistry - Process Audits	29 May 2009	29 May 2009	11 August 2009
SOP Review - In-life	08 June 2009	08 June 2009	11 August 2009
Validation of Procedure - In-life	08 June 2009	08 June 2009	11 August 2009
Protocol Review	10 June 2009	10 June 2009	11 August 2009
Protocol Amendment Review	10 June 2009	10 June 2009	11 August 2009
SOP Review - In-life	10 June 2009	10 June 2009	11 August 2009
Validation of Procedure - In-life	11 June 2009	11 June 2009	11 August 2009
Anchem Dose Data Anchem Dose Report - Report Review Anchem Dose Report Tabulation	28 July 2009 to 30 July 2009	30 July 2009	11 August 2009
Anchem Dose Data Final Report Review	22 March 2010 to 24 March 2010	24 March 2010	26 March 2010

In addition to the above-mentioned inspections, process based and/or routine facility inspections were also conducted during the course of this study. Any findings specific to this study from these inspections are reported with this QA Statement. All other observations and the dates of reports to PCS-MTL Management are retained on file according to PCS-MTL Quality Assurance Standard Operating Procedures.

31 Mar 2010
Date

Quality Assurance
Charles River Laboratories

3. SUMMARY

The purpose of this study was to determine the concentration of Perfluorohexanoic acid (PFH) ammonium salt in dose formulations from Charles River Laboratories Study No. UZS00009 titled “Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium Salt of Perfluorinated Hexanoic Acid) in Mice” by high performance liquid chromatography (HPLC).

The method of analysis, documented in Analytical Procedure AP.211053.SL.02 for concentration determination was previously validated under Study No. 211052. The method documented in Analytical Procedure AP.211053.PU.03, for bulk material purity and stability analysis was provided by the Sponsor.

The study samples analyzed were within the acceptance criteria of ±10% of their nominal concentrations and the stability samples were within acceptance criteria of ±10% of their initial concentration. For homogeneity, the relative standard deviation (RSD) for the formulation for each group was ≤ 5%.

The bulk material was analyzed for purity and stability, and the result was compared to the purity value stated on the Certificate of Analysis (CoA) and was deemed acceptable since the percentage difference was within ±10% of the value indicated in the Certificate of Analysis.

4. INTRODUCTION

A high performance liquid chromatographic (HPLC) method was used to determine the concentration of test article in dose formulations from Study No. UZS00009 titled "Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium Salt of Perfluorinated Hexanoic Acid) in Mice".

The method of analysis, documented in Analytical Procedure AP.211053.SL.02, was previously validated (Study No. 211052) and the method of analysis, documented in Analytical Procedure AP.211053.PU.03 was provided by the Sponsor.

For the work detailed in this report, the study initiation date was 20 May 2009 (the signature date of the protocol) and the completion date is the signature date of the final report. The experimental start date was 27 May 2009 and the experimental end date was 15 July 2009.

5. REFERENCE STANDARD AND VEHICLE

5.1. Reference Standard

Identity:	Perfluorohexanoic acid (PFH) C-1500N
Lot number:	7005
Purity:	47.4% (total purity)
Expiry date:	31 Jul 2010
Description:	Clear colourless liquid
Storage conditions:	Room temperature, light
Handling precautions:	As per the material safety data sheets
Supplier:	Charles River Pennsylvania

The reference standard characterization is the responsibility of the Sponsor who provided a Certificate of Analysis (Appendix 1) for inclusion in this study report. The expiry date was provided by the Sponsor in a written correspondence.

Details of identity, purity, storage conditions and handling precautions were supplied by the Sponsor. Remaining reference standard was used on subsequent studies for the Sponsor.

5.2. Vehicle

Identity:	Reverse osmosis deionized water
Storage conditions:	Room temperature

6. EXPERIMENTAL PROCEDURES

6.1. Standard Stock Solutions

Standard stock solutions of reference standard were prepared in diluent (acetonitrile:methanol:water (10:10:80, v/v/v) containing 0.1% (v/v) phosphoric acid) at a nominal concentration of 2.37 mg/mL.

6.2. Standard Solutions

Standard solutions of reference standard were prepared in diluent covering the nominal concentration range of 23.7 to 237 µg/mL.

6.3. Spiked Samples

Spiked samples were prepared in vehicle at nominal concentrations of 5.00 and 90.0 mg/mL. Each was diluted with diluent to give nominal concentrations of 80.0 and 180 µg/mL, respectively.

6.4. Study Samples

Formulation samples (top, middle and bottom) from study UZS00009 prepared on 25 May and sampled on 26 May 2009 were received at ambient temperature on 28 May 2009. Samples were received for concentration, homogeneity determination and stability assessments. The samples were stored at a target temperature of 4ºC for 1 day until analysis. Prior to analysis, the samples were brought to room temperature. The samples at nominal concentration of 7, 35 and 70 mg/mL were diluted with diluent to give injected concentrations within the range of the calibration curve.

Following the initial analysis (Day 0), formulation samples were stored at room temperature for approximately 10 days until analysis. Stability assessment was performed for the low, middle and high concentration samples. All samples were diluted with diluent to give injected concentrations within the range of the calibration curve.

For concentration analysis, the results were considered acceptable if the difference between the actual mean value and the targeted concentration was within ±10%. For homogeneity, the results were considered acceptable if the relative standard deviation (RSD) for the formulation calculated as the RSD for the grand mean of the average values for the top, middle and bottom locations was ≤ 5%. For stability analysis, results of the stability samples were to be within ±10% of the concentration of the initial stability samples to be acceptable.

6.5. Bulk Test Substance Stability

A 10 mL sample of the test substance (50% w/v) was received from study UZS00009 for stability assessment. The sample was shipped at ambient temperature on the 27 May 2009 and received at Charles River Montreal on the 28 May 2009. The sample was stored at room temperature and analyzed on 14 July 2009. The bulk substance was diluted 50 times with diluent (ultra pure water) to give a target concentration of 1% test substance. Stability was assessed by HPLC purity normalization and the result obtained was compared against the purity value stated in the Certificate of Analysis.

6.6. Analysis

The standard, blank, spiked sample and study sample solutions were analyzed by HPLC using the following conditions:

HPLC system: Agilent Technologies 1100 series
Data capture system: Waters Corporation Empower 2
Column: Zorbax Eclipse Plus C-18, 3.5 µm (100 x 2.1 mm id)
Column temperature: Set at 35°C
Mobile phase gradient elution: Eluant A: 20 mM sodium phosphate in water
Eluant B: 10 mM sodium perchlorate in acetonitrile

Time (min)	%B
0	10
8	70
8.1	10
15	10

Flow-rate: 0.350 mL/min
Ultra-violet detection wavelength: 210 nm (response time: 0.5 s)
Injection volume: 25 µL
Sample tray temperature: Set at 20ºC
Reference standard retention time: ~7.1 min

The blank and bulk substance solutions were analyzed for purity and stability using the following conditions:

HPLC system: Agilent Technologies 1100 series
Data capture system: Waters Corporation Empower 2
Column: TOSOH TSKGel ODS120T, (150 x 4.6 mm id)
Column temperature: Set at 40°C
Mobile phase gradient elution: Eluant A: acetonitrile
Eluant B: 0.6% perchloric acid in water

Time (min)	%B
0	50
10	50
15	10
20	10
20.1	50
25	50

Flow-rate: 1.00 mL/min
Ultra-violet detection wavelength: 210 nm
Injection volume: 20 µL
Sample tray temperature: Set at 20ºC
Reference standard retention time: ~4.0 min

6.7. System Suitability

For concentration determination, the reproducibility of the chromatographic system was determined by injecting a calibration standard solution, at a nominal concentration of 190 µg/mL in triplicate, at the beginning, throughout, and at the end of the chromatographic run.

For bulk substance stability, the reproducibility of the chromatographic system was determined by injecting a 1% test sample solution, in triplicate, at the beginning and at the end of the chromatographic run.

A coefficient of variation (CV) of ≤ 3% in peak area and a difference of ±10% between the average response for the standards (test sample solution) injected at the end and throughout the run, compared with those injected at the beginning were considered acceptable.

6.8. Data Collection and Statistical Methods

Data collection was performed using Empower 2, from Waters Corporation.

Statistical analyses included linear regression using Empower 2 and descriptive statistics such as arithmetic means and standard deviations, using Microsoft Excel (Version 2000/2003).

Tables were prepared from retrospective manual entry on computer (Microsoft Word, Version 2000/2003). The data presented in the tables were calculated from non-rounded values and may not be accurately reproduced from the individual data presented.

6.9. Quality Assurance

The Quality Assurance department of PCS-MTL undertook and documented inspections and process audits of the analytical laboratory during the study conduct, and audited the study report as well as the raw data. The Quality Assurance Statement is presented on Page 7.

6.10. Archives

All raw data and documents generated at PCS-MTL during this study, and the final report will be transferred to the scientific archives of the testing facility after dispatch of the final report.

7. PROTOCOL DEVIATIONS

As per study protocol, stability of the formulation samples was to be assessed at low and high concentration. However, stability was assessed at low, intermediate and high concentrations. In addition, per the protocol, the study was to comply with regulations from the Ministry of Agriculture, Forestry and Fisheries; however, the analytical portion of the study did not comply with it since the test site is not accredited by this organization. In the Principal Investigator's opinion, these deviations are considered to have no impact on the outcome of the study as the work conducted was compliant to the other regulation agencies mentioned above, and that the work was reviewed by the Quality Assurance Unit and that all results were within acceptance criteria

8. RESULTS AND DISCUSSION

Representative chromatograms are presented in Figure 1, Figure 2, Figure 3, Figure 4, Figure 5 and Figure 6.

8.1. System Suitability

The CV for the calibration standards was ≤ 3%, and the difference between the average response for the standards injected at the end and throughout the run, compared with those injected at the beginning was within ±10%. Acceptance criteria with respect to system suitability were met.

For bulk substance stability, the CV for 1% test sample solution was ≤ 3%, and the difference between the average responses for the test sample solutions injected at the end, compared with those injected at the beginning was within ±10%.

8.2. Study Samples

All study samples analyzed for concentration were within the acceptance criteria of ±10% of their target values. For homogeneity, the relative standard deviation of the grand mean for all locations was ≤ 5% for all groups.

For each group, all stability samples analyzed were within ±10% of the initial concentrations analysis. Analysis of the bulk test material is on-going and results will be added to the revised draft report. Results are presented in Table 1 and Table 2.

8.3. Bulk Test Substance Stability

Stability of the bulk substance was assessed and the purity was determined to be 99.9%. The difference between the purity value obtained, when compared with the purity value indicated on the Certificate of Analysis was 7.0%. Results are presented in Table 3.

9. CONCLUSION

The dose formulations were within specification. Homogeneity results show that the formulation technique used produces homogenous preparations. Stability of the formulated reference standard was demonstrated at room temperature for 10 days. In addition, purity and stability of the bulk reference material was assessed following 48 days of storage at room temperature and results were deemed acceptable since the percentage difference was within ±10% of the value indicated in the Certificate of Analysis.

10. REPORT REVIEW AND APPROVAL SIGNATURE

This report has been reviewed by:

_____ 31 Mar. 2010
Date

Scientific Director
Analytical Chemistry, Laboratory Sciences
Charles River Laboratories

Table 1 Study Samples - Concentration and Homogeneity

Sampling date	Group	Nominal concentration (mg/mL)	Sampling location	Measured concentration		Homogeneity (RSD)
				(mg/mL)	(Percent of nominal)	
26 May 2009	1	7	Top	7.06	101	0.1
				7.07	101	
			Middle	7.05	101	
				7.06	101	
			Bottom	7.04	101	
				7.08	101	
			Mean	7.06	101	
	2	35	Top	34.7	99.3	0.0
				34.7	99.2	
			Middle	34.7	99.1	
				34.8	99.4	
			Bottom	34.7	99.2	
				34.7	99.2	
			Mean	34.7	99.2	
	3	70	Top	70.1	100	0.0
				70.2	100	
			Middle	70.3	100	
				69.6	99.4	
			Bottom	70.1	100	
				70.1	100	
			Mean	70	100	

LLOQ - lower limit of quantitation (5.00 mg/mL)

Table 2 Study Samples - Stability Assessment Following 10 Days Storage at Room Temperature

Nominal concentration (mg/mL)	Group ID	Measured concentration fresh (mg/mL)	Mean measured concentration fresh (mg/mL)	Measured concentration after storage (mg/mL)	Mean measured concentration after storage (mg/mL)	Mean difference fresh vs stored (%)
7	Top	7.06	7.06	6.98	7.01	-0.7
		7.07		7.03		
	Middle	7.05		7.01		
		7.06		7.02		
	Bottom	7.04		7.02		
		7.08		7.01		
35	Top	34.7	34.7	34.7	34.7	0.0
		34.7		34.7		
	Middle	34.7		34.7		
		34.8		34.7		
	Bottom	34.7		34.7		
		34.7		34.7		
70	Top	70.1	70.0	69.7	69.4	-0.9
		70.2		69.3		
	Middle	70.3		69.5		
		69.6		69.4		
	Bottom	70.1		69.2		
		70.1		69.4		

Table 3 Bulk Substance Stability (48 Days Storage at Room Temperature)

Bulk substance assessed purity (%)	Bulk substance impurity (%)	Bulk material CoA purity (%)	Bulk material CoA total impurity (%)	Percent difference [a]
99.9	0.1	93.4	6.6	7.0

a assessed purity is compared with the purity stated on the CoA

Figure 1 Representative Standard Chromatogram (Nominal Concentration: 190 µg/mL)

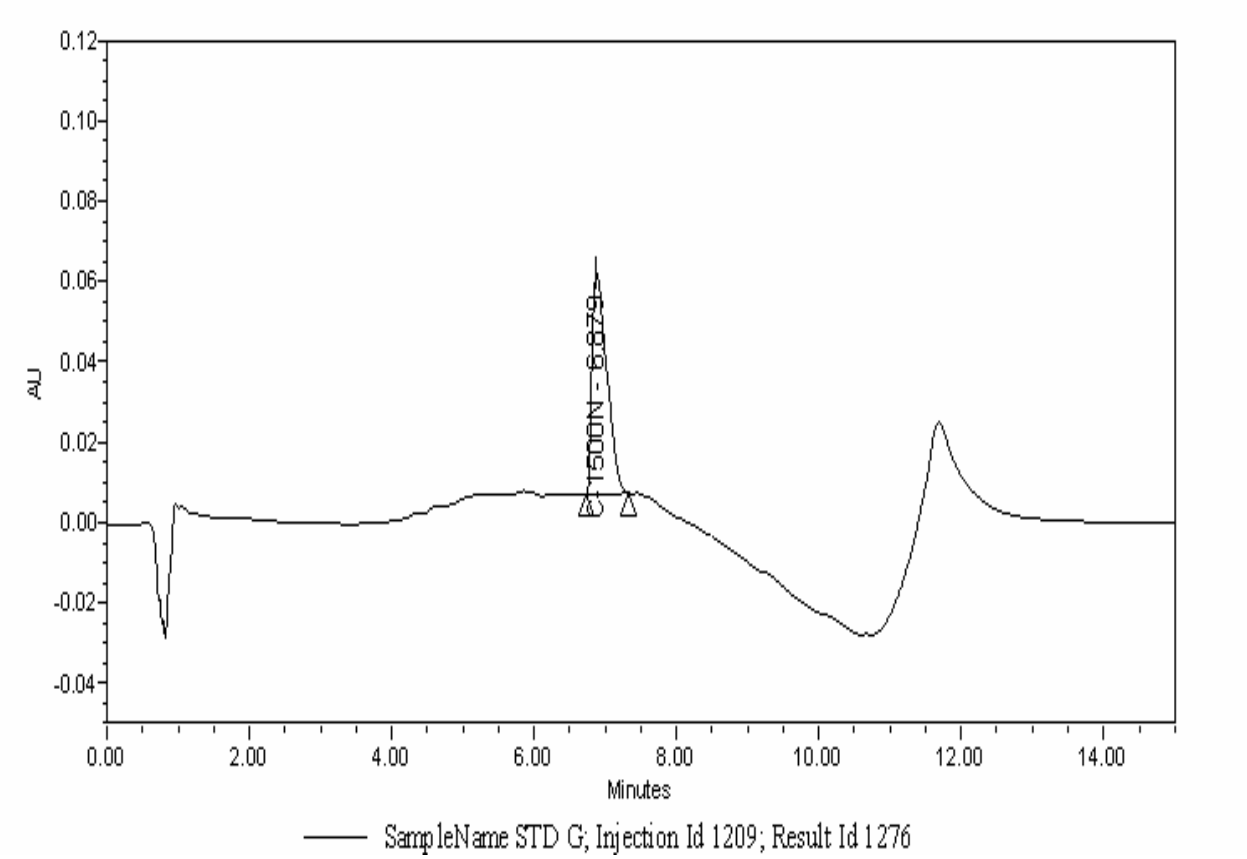


Figure 2 Representative Blank Vehicle

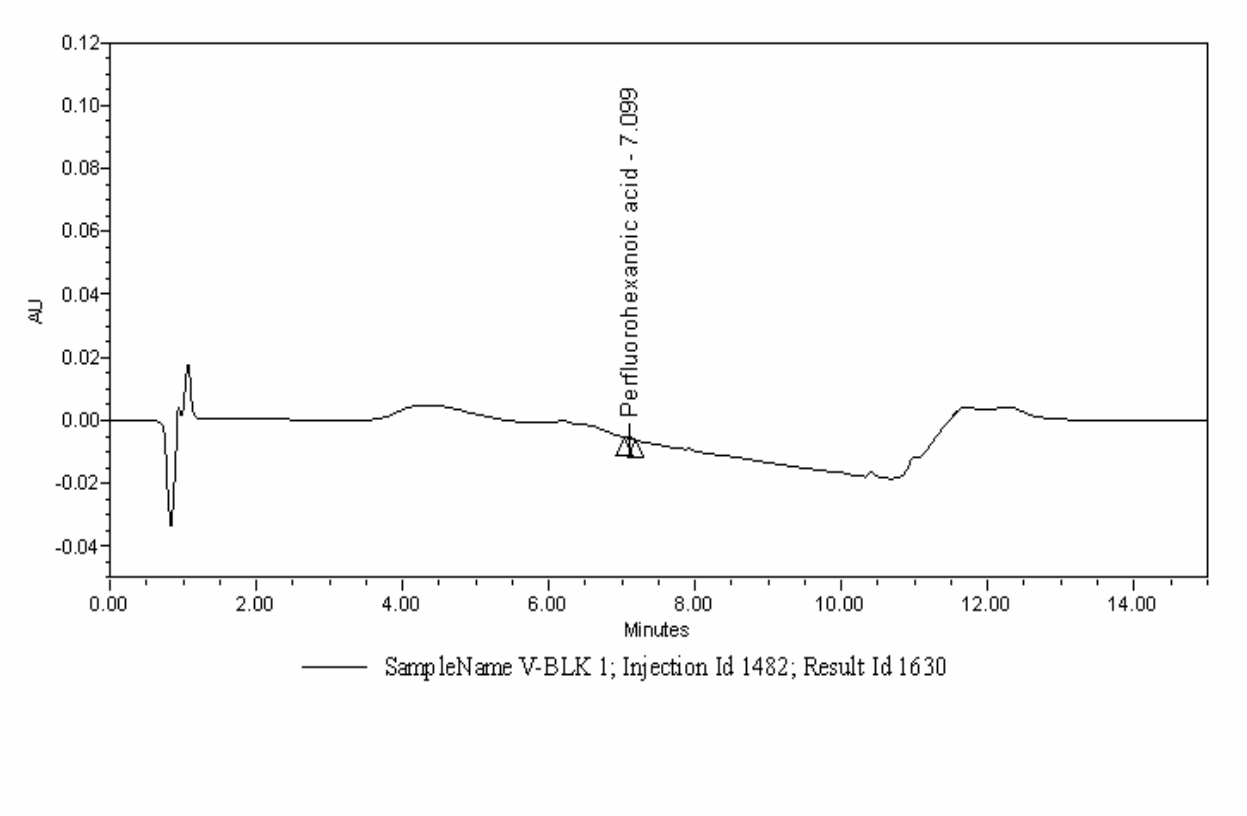


Figure 3 **Representative Sample Chromatogram (Group 1, Top, Sampling Date: 26 May 2009, Nominal Concentration: 7 mg/mL; Nominal Injected Concentration: 105 µg/mL)**

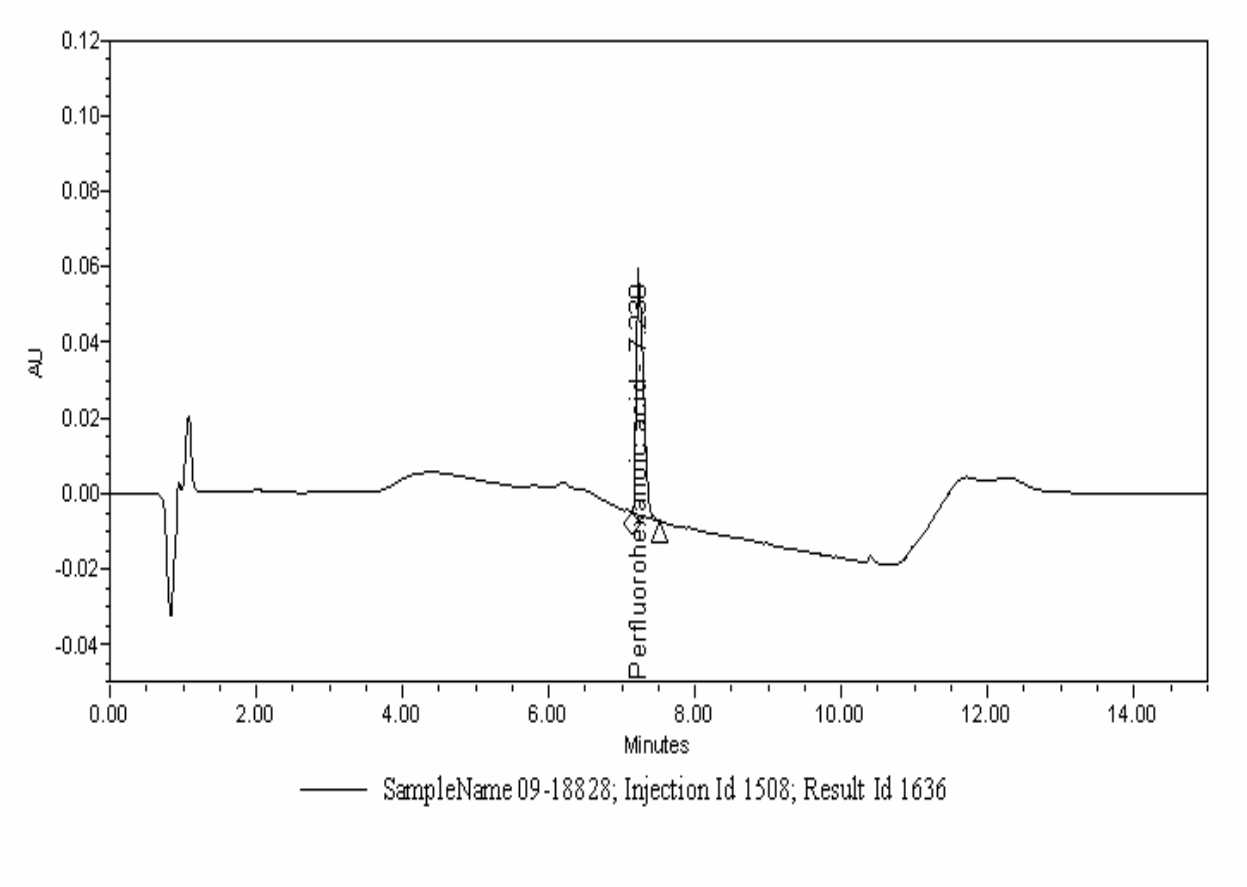


Figure 4 Representative Bulk Substance Sample (Full Scale)

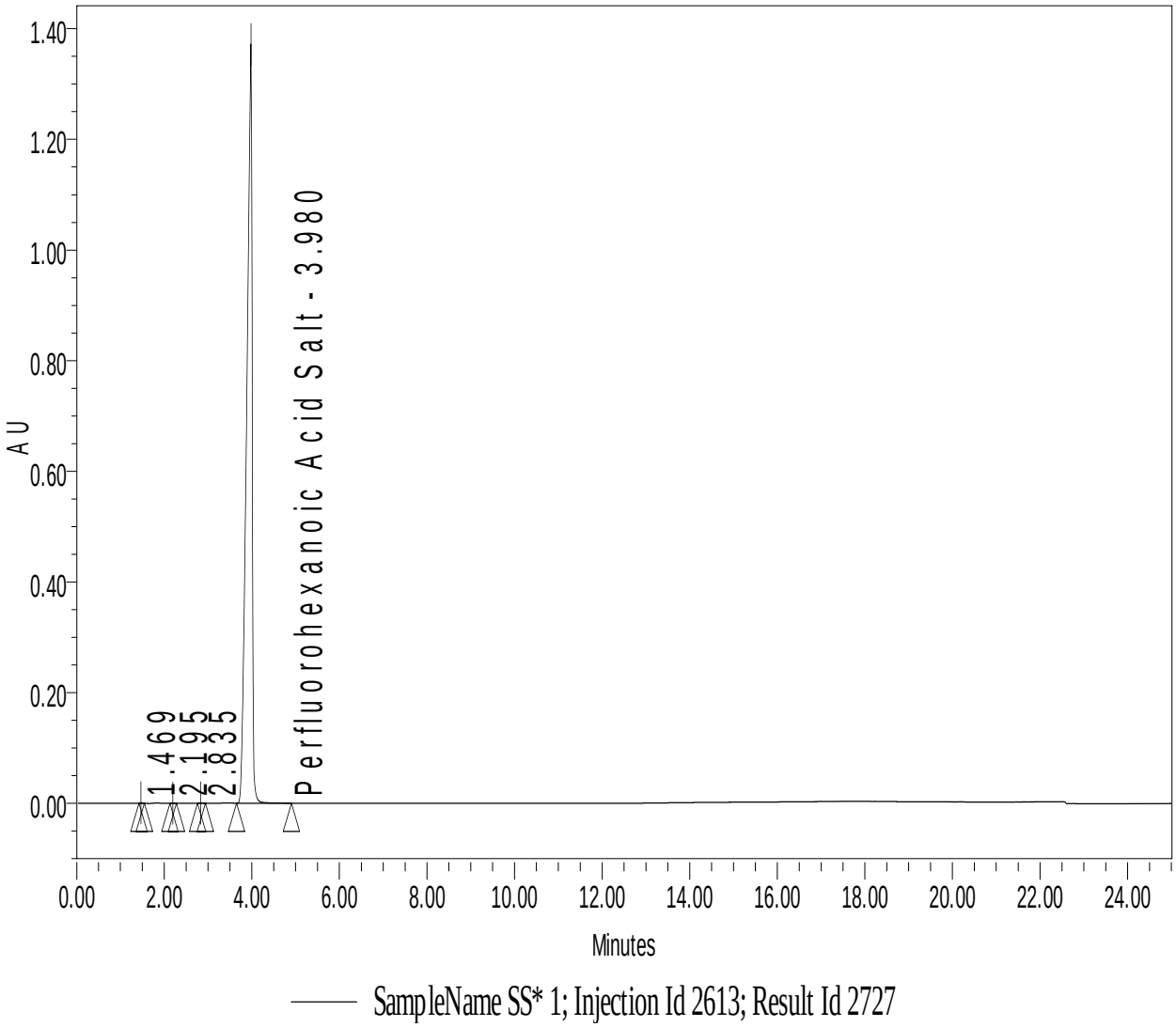
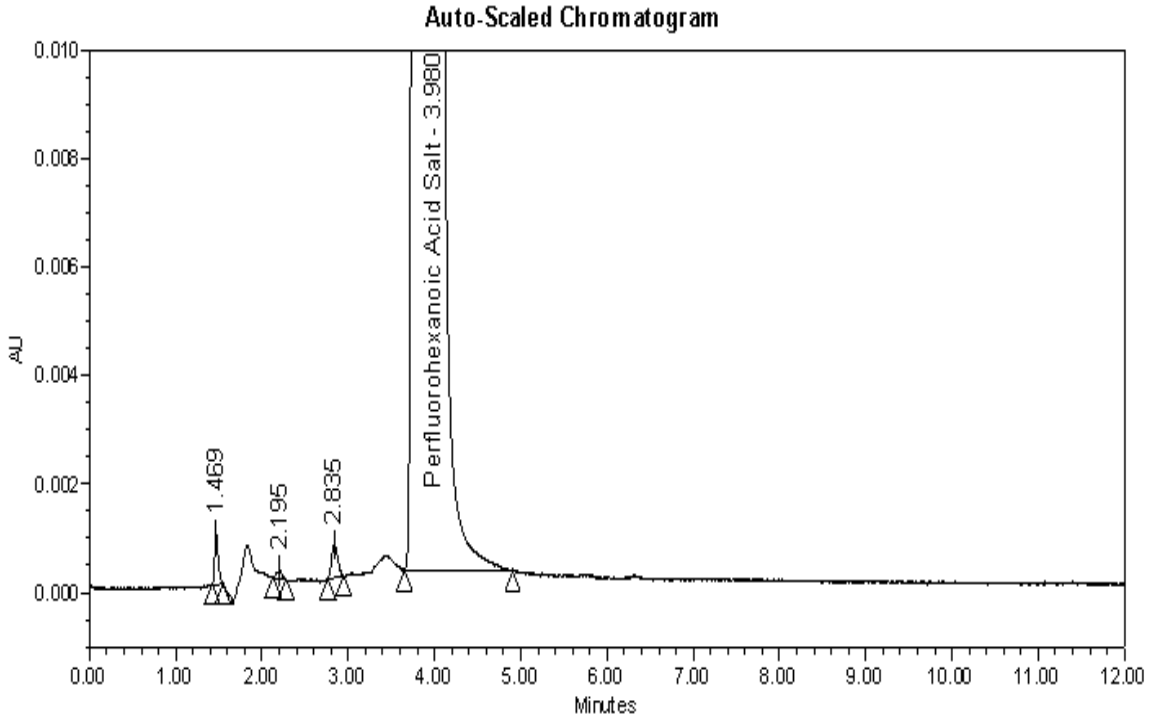


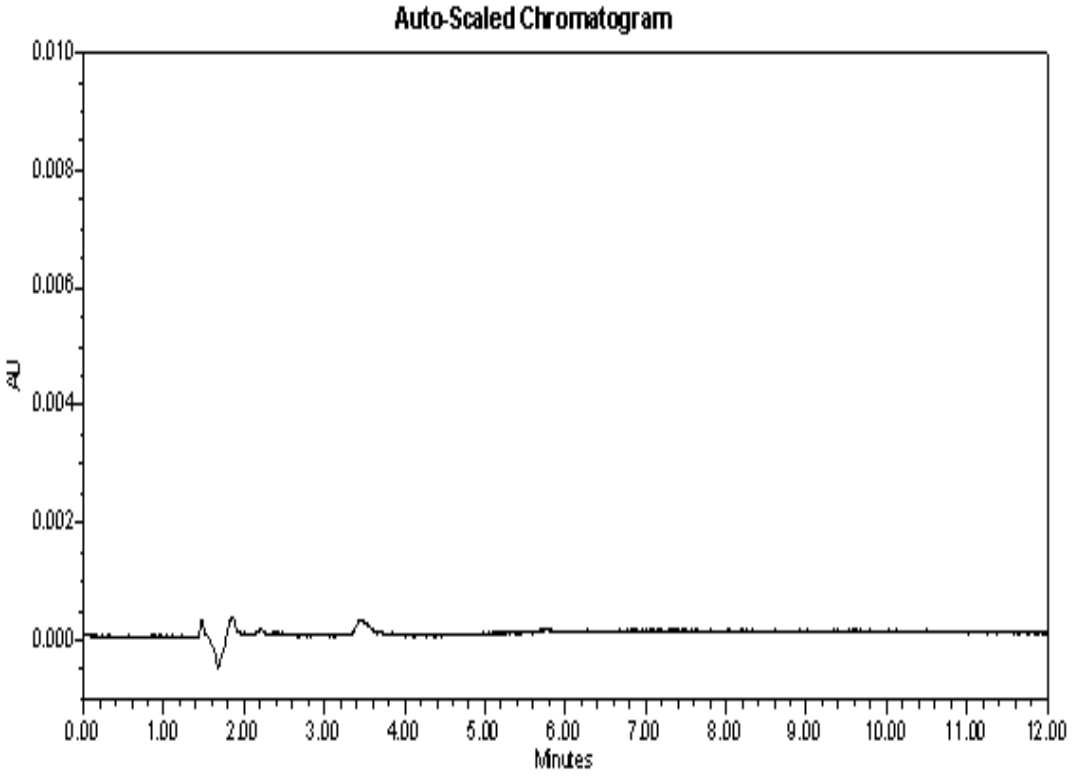
Figure 5 Representative Bulk Substance Sample (Auto-scaled)



Peak Results

	Name	SMP_Name	Injection Id	Result Id	% Area	Area	RT
1		SS* 1	2613	2727	0.02	2646	1.469
2		SS* 1	2613	2727	0.01	636	2.195
3		SS* 1	2613	2727	0.03	3118	2.835
4	Perfluorohexanoic Acid Salt	SS* 1	2613	2727	99.95	11639275	3.980

Figure 6 Representative Blank Sample (Auto-scaled)



Peak Results

	Name	SMP_Name	Injection Id	Result Id	% Area	Area	RT
1	Perfluorohexanoic Acid Salt	BLK*	2593	2655			3.980

Appendix 1 Certificate of Analysis

Certificate of Analysis

Daikin Industries,LTD.

Name of Sample	*PFH Ammonium Salt（C-1500N）*
Lot.	*7005*
Date of Analysis	*May 14, 2009*
Purify	*47.4% (Effective component in Water)*
	**50.8*0.934%=47.4%*

COMPOSITION

identity		*Conc.*
#1	*Ammonium Perfluorohexanoate CAS RN. 21615-47-4*	*93.4%*
#2	*Unknown*	*6.6%*
	Total	*100%*

Analysis system (HPLC)

Equipment	*: Waters Alliance2695*
Detector	*: Waters 2487UV*
Detection wavelength	*: 210nm*

Analysis condition

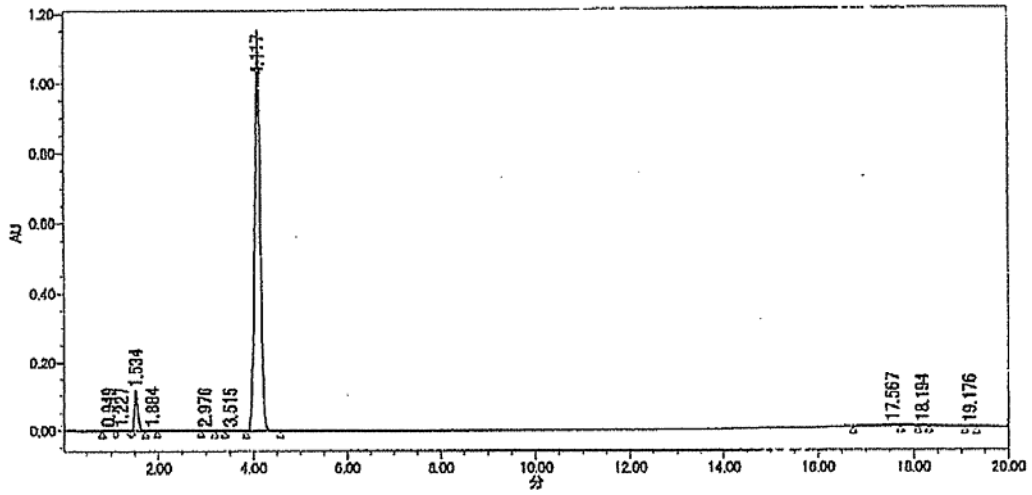
Column	*: TOSOH TSKGel ODS120T 4.6mm×150mm*
Temp.	*: 40 ℃*
Mobile phase	*: A=acetonitrile 、B=Solution of 0.6% perchloric acid in water*
Gradient	*: A:B=50:50(mass%) (0-10min.) →90:10(mass%) (15-20min.)*
Injection volume	*: 20μL*
Injection Concentration	*: 1%（dilute 50times with water)*

Chemical R&D Center
Unidyne Group
Senior Researcher

SIGNATURE
DATE : May 18, 2009

Analysis

サンプル名：	C1500N	分析担当者：	System
サンプルの種類：	未知試料	分析日：	2009/05/14 11:49:44
バイアル：	82	取り込みメソッドセット：	090514S
注入#：	1	解析日：	2009/05/14 13:55:17
注入量：	20.00 μl	解析メソッド：	C1500N
分析時間：	20.00 分	チャンネル名：	2487チャンネル 1
サンプルセット名：		解析チャンネルの説明：	



	成分名	Retention Time(min.)	Area (μVsec.)	Area (%)	Height (μV)		
1		0.949	17634	0.18	2554		
2		1.227	20551	0.20	1927		
3		1.534	574660	5.71	116134		
4		1.884	5543	0.06	710		
5		2.976	2424	0.02	414		
6		3.515	4940	0.05	361		
7		4.117	9390042	93.38	1144218		
8		17.567	29475	0.29	984		
9		18.194	6956	0.07	1098		
10		19.176	3881	0.04	592		

1/1 ページ

Appendix 2 Principal Investigator Acknowledgment Form

Principal Investigator Acknowledgment Form

Testing Facility Study No.: UZS00009

INSTRUCTIONS: This acknowledgement form is to be signed, dated and returned to the Testing Facility Study Director prior to the phase(s) of the study being conducted under the responsibility of the Principal Investigator (PI) at the PI's site.

Principal Investigator Name: Kapinga Mbuy, BSc (Dose Formulation Analysis)

The undersigned certifies hereby that the study phase(s) conducted at the PI site will be performed in compliance with the Principles of Good Laboratory Practices and in accordance with the study protocol.

29 May 2009
Date

Please return to:
Name:
Charles River Laboratories
Preclinical Services, Pennsylvania
905 Sheehy Drive, Building 905
Horsham, PA 19044

FAX: 215-443-8587 E-Mail:

05.19.06 PIAF1-01 32D.7

APPENDIX 4 - PHARMACOKINETIC ANALYSIS REPORT

charles river

FINAL PHARMACOKINETIC REPORT

Test Site Ref. No. 480173
Testing Facility Study No. UZS00009

Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium Salt of Perflourinated Hexanoic Acid) in Mice

TEST SITE:
Charles River Laboratories Preclinical Services Montreal
22022 Transcanadienne
Senneville, Quebec
Canada H9X 3R3

TESTING FACILITY:
Charles River Laboratories Preclinical Services
905 Sheehy Drive, Building A
Horsham, PA 19044
USA

SPONSOR:
Daikin Industries, LTD
Chemical Division
Umeda Center Building
4-12 Nakazaki-Nishi, 2-chrome
Kita-ku, Osaka 530-8323
Japan

22 April 2010

Page 1 of 25

TABLE OF CONTENTS

LIST OF TABLES 3
LIST OF FIGURES 4
LIST OF APPENDICES 5
GLOSSARY OF TERMS 6
1. COMPLIANCE STATEMENT 7
2. QUALITY ASSURANCE STATEMENT 8
3. INTRODUCTION 9
3.1. Objectives 9
4. EXPERIMENTAL DESIGN 9
4.1. Sampling Schedule 9
5. METHODS 10
5.1. Pharmacokinetic Procedure 10
5.2. Statistical Analysis 10
6. QUALITY ASSURANCE 11
7. ARCHIVES 11
8. RESULTS AND DISCUSSION 11
8.1. Serum Concentration Observations 11
8.2. Toxicokinetic Evaluation 11
9. SUMMARY AND CONCLUSION 12
10. REPORT REVIEW AND APPROVAL SIGNATURE 13
11. REFERENCES 14

LIST OF TABLES

Table 1.1-1.3 Concentrations of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt.....15

Table 2 Pharmacokinetic Observed and Secondary Parameters of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt.....18

Table 3 Pharmacokinetic Exposure Parameters of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt.....19

Table 4 Dose Proportionality of PFHxA Cmax and AUC(0-inf) in Female Crl:CD1(ICR) Mice Serum Relative to Ascending Dose Level of PFH Ammonium Salt Following Oral Gavage.....20

LIST OF FIGURES

Figure 1 Pharmacokinetic Profiles of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt.....21

Figure 2 Comparison of PFHxA Dose Normalized Exposure (Cmax/Dose and AUC(0-inf)/Dose) in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt.....22

Figure 3 Comparison of PFHxA Exposure (Cmax and AUC(0-inf)) in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt.....23

LIST OF APPENDICES

Appendix 1 Data Capture/Entry Methods Used in Report 24

GLOSSARY OF TERMS

Tmax	The time of maximum observed concentration after dosing
Tlast	The time of last quantifiable concentration after dosing
Kel	Apparent terminal elimination rate constant
Rsq	Coefficient of determination for the terminal elimination phase regression model
T1/2	Apparent terminal elimination half-life
Cmax	The maximum observed concentration after dosing
AUC(0-t)	The area under the concentration versus time curve from time zero to Tlast
AUC(0-inf)	The area under the concentration versus time curve from time zero to infinity
AUC%extrap	Extrapolated area under the concentration versus time curve expressed as percent of the total AUC(0-inf)
Cmax/D	The maximum observed concentration after dosing, normalized for the dose administered
AUC(0-inf)/D	The area under the drug concentration versus time curve from time zero to infinity, normalized for the dose administered

Additional parameters were automatically generated by the software but were not reported.

1. COMPLIANCE STATEMENT

This analysis was conducted at Charles River Laboratories Preclinical Services Montreal (PCS-MTL), 22022 Transcanadienne, Senneville, Quebec, Canada, H9X 3R3, started on 04 November 2009 and was completed on 04 November 2009.

These pharmacokinetic analyses were performed in accordance with:

- PCS-MTL Standard Operating Procedures
- The United States Environmental Protection Agency, Code of Federal Regulations, Title 40, Parts 160 and 792: Good Laboratory Practice Standards
- The Organisation for Economic Co-operation and Development (OECD) Principles on Good Laboratory Practice [C (97) 186/Final] (1998)

Due to the fact that the test site is not accredited by this agency, this portion of the study is not compliant with the Good Laboratory Practice Standards for Toxicological Studies on
istry of Agriculture, Forestry and Fisheries.

22 Apr 2010
Date

Principal Investigator
Senior Research Scientist
Pharmacokinetics
Charles River Laboratories

2. QUALITY ASSURANCE STATEMENT

In compliance with the Good Laboratory Practice Regulations, Reference No. 480173 has been audited. The data presented in the final report accurately represent the data collected during the conduct of the study.

Phase or Segment Audited	Date of Inspection	Dates of Reports to Test Site Management and Principal Investigator	Dates of Reports to Testing Facility Management/ Study Director & Lead QA
Protocol Review	15 June 2009	15 June 2009	10 December 2009
Protocol Amendment Review	15 June 2009	15 June 2009	10 December 2009
Protocol Amendment Review	30 November 2009	04 December 2009	10 December 2009
PK/TK - Data Review Method of Data Capture - Tabulated Data PK/TK - Tabulated Data PK/TK - Report Review	30 November 2009 to 04 December 2009	04 December 2009	10 December 2009
Final Report Review	05 April 2010 to 13 April 2010	13 April 2010	20 April 2010

_____ 22-Apr-2010
Date

Inspector
Quality Assurance
Charles River Laboratories

3. INTRODUCTION

Study No. UZS00009 was undertaken to evaluate the toxicity of acute exposure of Crl:CD1(ICR) female mice to PFH Ammonium Salt (Ammonium salt of Perflourinated Hexanoic Acid, PFHxA). Pharmacokinetics were included as part of this assessment. In addition, the study was extended in order to attain the appropriate range for validation of the assay in the liver. This information may be used in determining dosage levels for the segment II/III toxicity study in mice.

Administration of the test and control substances, and blood collection and processing for this study were performed at Charles River Laboratories Preclinical Services Pennsylvania under GLP conditions. The PFHxA serum concentration data analyzed in this report were produced at PCS-MTL under GLP conditions, using a validated LC-MS/MS method under Reference No. 141660.

3.1. Objectives

The primary objective of this pharmacokinetic (PK) analysis was to describe the systemic exposure of PFHxA achieved in female Crl:CD1(ICR) mice when administered once by oral gavage, and its relationship to dose level (ICH S3A, 1995).

4. EXPERIMENTAL DESIGN

PFHxA Ammonium Salt was administered once by oral gavage to female Crl:CD1(ICR) mice. The control substance used for this study was reverse osmosis deionized water. Text Table 1 illustrates the experimental design of the study.

Text Table 1

Dosage Group	Batch Number	Dose Level (mg/kg)	Concentration (mg/mL)	Dosage Volume (mL/kg)	Number of Mice
I	B-UZS00009-A	35	7	5	21
II	B-UZS00009-B	175	35	5	21
III	B-UZS00009-C	350	70	5	21

The test substance was considered 95% by weight of PFHxA for dosage calculations

4.1. Sampling Schedule

Blood samples were collected from PK animals, into tubes devoid of anticoagulant, according to the schedule in Text Table 2, prior to being processed to serum.

Text Table 2

Dosage Group	Number of Animals/Group	Toxicokinetic Time-point (Hours Post Dose)						
		0[a]	0.5	2	4	6	8	24
I to III	3	X						
	3		X					
	3			X				
	3				X			
	3					X		
	3						X	
	3							X

a samples collected prior to dosing
X = sample collected

5. METHODS

5.1. Pharmacokinetic Procedure

Serum concentration data were obtained from 3 animals at each time point, and mean values were used to generate a composite PK profile. Serum concentration values below the lower limit of quantitation (LLOQ) of the assay (1 µg/mL) were assigned a value of zero (µg/mL) for the purpose of mean calculation. If the resulting mean concentration returned a value below the LLOQ, it was treated as an absent sample for PK analysis. The PK profiles were characterized by non-compartmental analysis of PFHxA serum concentration data with targeted sampling time-points using validated computer software (WinNonlin, version 3.2, Pharsight Corp., Mountain View, California, USA). A model was selected based on the extravascular route of administration and the serum matrix.

The area under the test article serum concentration vs. time curve (AUC) was calculated using the linear trapezoidal method (linear interpolation). When practical, the terminal elimination phase of the PK profiles was identified based on the line of best fit using at least the final three observed concentration values. The slope of the terminal elimination phase was calculated using log-linear regression using the unweighted concentration data. PK parameters describing the systemic exposure of the test article in the test system were estimated from observed (rather than predicted) serum concentration values, the dosing regimen, the AUC, and the terminal elimination phase rate constant (Kel) for each group.

5.2. Statistical Analysis

Where applicable, numerical data obtained during the conduct of the study were subjected to calculation of descriptive statistics (mean and standard deviation) in Microsoft Excel, 2000/2003.

6. QUALITY ASSURANCE

The Quality Assurance department of PCS-MTL documented audits of the PK data and report. Data audits were conducted according to PCS-MTL standard operating procedures.
Any findings of QA inspections were reported to the Principal Investigator, the Study Director, the Testing Facility Management and the Test Site Management.

7. ARCHIVES

All raw data, copy of protocol and amendments, and documents generated at PCS-MTL during this analysis, including the final PK contributing report, will be transferred to the scientific archives of PCS-MTL at or prior to signature of the report, and will be kept for a period of one year following dispatch of the final PK contributing report. Subsequently, storage details will be documented in the raw data.

8. RESULTS AND DISCUSSION

8.1. Serum Concentration Observations
(Table 1.1 to Table 1.3 and Figure 1)

The serum concentrations of PFHxA were not quantifiable in predose samples in any of the groups. In the 35 mg/kg dose group, no PFHxA could be quantified after 6 hours. In the 175 and 350 mg/kg dose groups, the mean concentration of PFHxA was below the LLOQ at 24 hours post dose, but one individual animal in each group had quantifiable levels at 24 hours.

8.2. Toxicokinetic Evaluation
(Table 2, Table 3, Table 4 and Figure 1 to Figure 3)

The maximal serum concentrations (Cmax) of PFHxA were observed at the first sampling time point (30 minutes post dose), indicating that the compound was rapidly absorbed after oral administration. After Cmax was reached, serum concentrations of PFHxA decreased rapidly. At the 35 mg/kg dose level, the decline in PFHxA serum concentration was mono-exponential. At the 175 mg/kg and 350 mg/kg dose levels, the decline was multi-exponential, with a plateau in concentrations occurring between 0.5 and 2 hours in the 175 mg/kg dose group and between 2 and 4 hours in the 350 mg/kg dose group. The terminal elimination phases were nonetheless similar among the three dose groups, with terminal elimination half-lives ranging from 0.889 to 1.24 hours. Exposure increased with increase in dose level. The increase in Cmax (from 96.6 to 454 µg/mL) was lower than proportional, whereas the increase in AUC(0-inf) (from 178 to 1893 h*µg/mL) was proportional, to the increase in dose from 35 to 350 mg/kg.

9. SUMMARY AND CONCLUSION

The pharmacokinetics of PFHxA were characterized in female Crl:CD1(ICR) mice when administered once by oral gavage at dose levels of 35, 175 and 350 mg/kg. PFHxA was rapidly absorbed (Cmax reached within 30 minutes) and in general was not quantifiable at 24 hours after dose administration. The terminal elimination half-life of PFHxA ranged from 0.889 to 1.24 hours, and was dose-independent. The increase in Cmax (from 96.6 to 454 µg/mL) was lower than proportional, whereas the increase in AUC(0-inf) (from 178 to 1893 h*µg/mL) was proportional, to the increase in dose from 35 to 350 mg/kg.

10. REPORT REVIEW AND APPROVAL SIGNATURE

This report has been reviewed and approved by:

_____ 22 April 2010
Date

Scientific Director
Drug Metabolism and Pharmacokinetics *In Vivo*
Charles River Laboratories

11. REFERENCES

International Conference on Harmonization (ICH) Guideline S3A: Note for Guidance on Toxicokinetics: The Assessment of Systemic Exposure in Toxicity Studies. United States Food and Drug Administration Federal Register, Vol. 60, No. 40, 01 March 1995, pages 11264-11268.

Table 1.1 Concentrations of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt

	Group I: 35 mg/kg - Concentration (µg/mL)						
	Nominal Time						
Animal No.	Predose	0.5 h	2 h	4 h	6 h	8 h	24 h
3000	<LLOQ	-	-	-	-	-	-
3001	<LLOQ	-	-	-	-	-	-
3002	<LLOQ	-	-	-	-	-	-
1204	-	132	-	-	-	-	-
1205	-	65.3	-	-	-	-	-
1206	-	92.8	-	-	-	-	-
1207	-	-	36.0	-	-	-	-
1208	-	-	22.4	-	-	-	-
1209	-	-	49.7	-	-	-	-
1210	-	-	-	6.73	-	-	-
1211	-	-	-	12.1	-	-	-
1212	-	-	-	3.90	-	-	-
1213	-	-	-	-	3.25	-	-
1214	-	-	-	-	<LLOQ	-	-
1215	-	-	-	-	1.53	-	-
1216	-	-	-	-	-	<LLOQ	-
1217	-	-	-	-	-	<LLOQ	-
1218	-	-	-	-	-	<LLOQ	-
1219	-	-	-	-	-	-	<LLOQ
1220	-	-	-	-	-	-	<LLOQ
1221	-	-	-	-	-	-	<LLOQ
Mean	<LLOQ	96.6	36.0	7.58	1.59	<LLOQ	<LLOQ
SD	n/a	33.4	13.6	4.17	1.62	n/a	n/a

<LLOQ = Below the lower limit of quantitation (LLOQ = 1 µg/mL). The <LLOQ concentrations were assigned a value of zero for mean calculation. Mean values <LLOQ were not reported.

n/a = Not applicable.

- Sample not collected.

Table 1.2 Concentrations of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt

	Group II: 175 mg/kg - Concentration (µg/mL)						
	Nominal Time						
Animal No.	Predose	0.5 h	2 h	4 h	6 h	8 h	24 h
1222	<LLOQ	-	-	-	-	-	-
1223	<LLOQ	-	-	-	-	-	-
1224	<LLOQ	-	-	-	-	-	-
1225	-	364	-	-	-	-	-
1226	-	288	-	-	-	-	-
1227	-	338	-	-	-	-	-
1228	-	-	317	-	-	-	-
1229	-	-	360	-	-	-	-
1230	-	-	264	-	-	-	-
1231	-	-	-	69.9	-	-	-
1232	-	-	-	118	-	-	-
1233	-	-	-	101	-	-	-
1234	-	-	-	-	39.5	-	-
1235	-	-	-	-	8.37	-	-
1236	-	-	-	-	8.31	-	-
1237	-	-	-	-	-	2.11	-
1238	-	-	-	-	-	2.38	-
1239	-	-	-	-	-	34.4	-
1240	-	-	-	-	-	-	<LLOQ
1241	-	-	-	-	-	-	2.02
1242	-	-	-	-	-	-	<LLOQ
Mean	<LLOQ	330	313	96.2	18.7	13.0	<LLOQ
SD	n/a	38.7	47.8	24.4	18.0	18.6	n/a

<LLOQ = Below the lower limit of quantitation (LLOQ = 1 µg/mL). The <LLOQ concentrations were assigned a value of zero for mean calculation. Mean values <LLOQ were not reported.

n/a = Not applicable.

- Sample not collected.

Table 1.3 Concentrations of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt

	Group III: 350 mg/kg - Concentration (µg/mL)						
	Nominal Time						
Animal No.	Predose	0.5 h	2 h	4 h	6 h	8 h	24 h
1243	<LLOQ	-	-	-	-	-	-
1244	<LLOQ	-	-	-	-	-	-
1245	<LLOQ	-	-	-	-	-	-
1246	-	381	-	-	-	-	-
1247	-	509	-	-	-	-	-
1248	-	473	-	-	-	-	-
1249	-	-	440	-	-	-	-
1250	-	-	124	-	-	-	-
1251	-	-	301	-	-	-	-
1252	-	-	-	317	-	-	-
1253	-	-	-	332	-	-	-
1254	-	-	-	229	-	-	-
1255	-	-	-	-	199	-	-
1256	-	-	-	-	103	-	-
1257	-	-	-	-	168	-	-
1258	-	-	-	-	-	24.1	-
1259	-	-	-	-	-	12.2	-
1260	-	-	-	-	-	7.45	-
1261	-	-	-	-	-	-	2.68
1262	-	-	-	-	-	-	<LLOQ
1263	-	-	-	-	-	-	<LLOQ
Mean	<LLOQ	454	288	293	157	14.6	<LLOQ
SD	n/a	66.0	158	55.6	48.9	8.59	n/a

<LLOQ = Below the lower limit of quantitation (LLOQ = 1 µg/mL). The <LLOQ concentrations were assigned a value of zero for mean calculation. Mean values <LLOQ were not reported.

n/a = Not applicable.

- Sample not collected.

Table 2 **Pharmacokinetic Observed and Secondary Parameters of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt**

Group No.	Dose Level (mg/kg)	Tmax (h)	Tlast (h)	Kel (1/h)	Rsq	T1/2 (h)
I	35	0.50	6.00	0.780	1.00	0.889
II	175	0.50	8.00	0.560	0.952	1.24
III	350	0.50	8.00	0.750	0.898	0.924

Table 3 **Pharmacokinetic Exposure Parameters of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt**

Group No.	Dose Level (mg/kg)	Cmax (µg/mL)	AUC(0-t) (µg•h/mL)	AUC(0-inf) (µg•h/mL)	AUC%extrap (tlast-inf)	Cmax/ Dose	AUC(0-inf)/ Dose
I	35	96.6	176	178	1.14	2.76	5.10
II	175	330	1121	1144	2.03	1.88	6.54
III	350	454	1873	1893	1.03	1.30	5.41

Table 4 **Dose Proportionality of PFHxA Cmax and AUC(0-inf) in Female Crl:CD1(ICR) Mice Serum Relative to Ascending Dose Level of PFH Ammonium Salt Following Oral Gavage**

Group No.	Dose Level (mg/kg)	Fold Increase	Cmax (µg/mL)	Fold Increase	AUC(0-inf) (µg•h/mL)	Fold Increase
I	35	1.00	96.6	1.00	178	1.00
II	175	5.00	330	3.41	1144	6.41
III	350	2.00	454	1.38	1893	1.65

Figure 1 Pharmacokinetic Profiles of PFHxA in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt

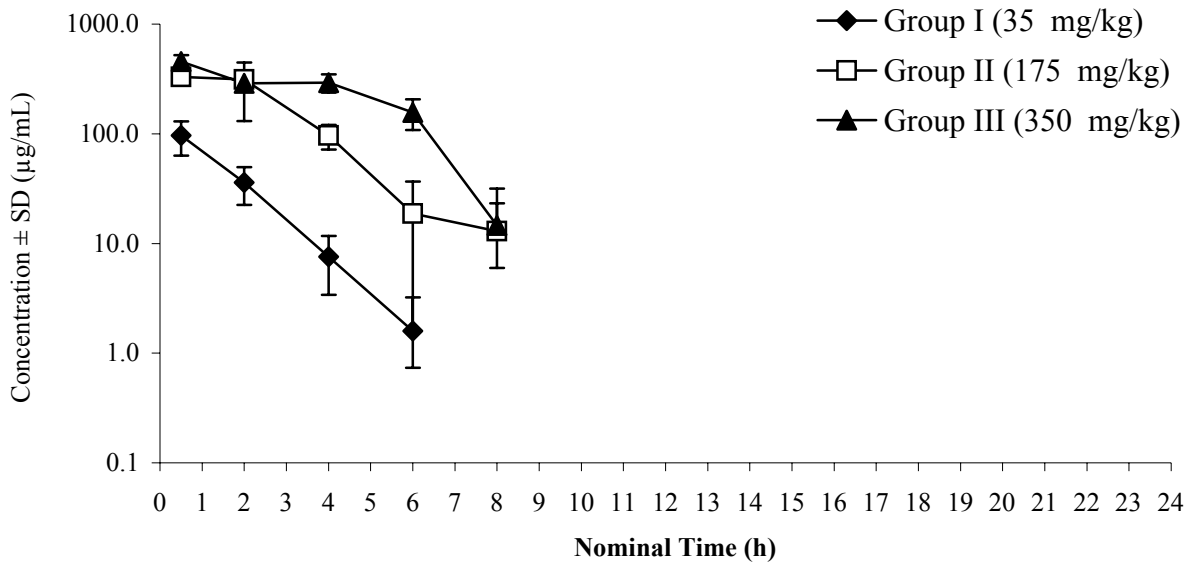
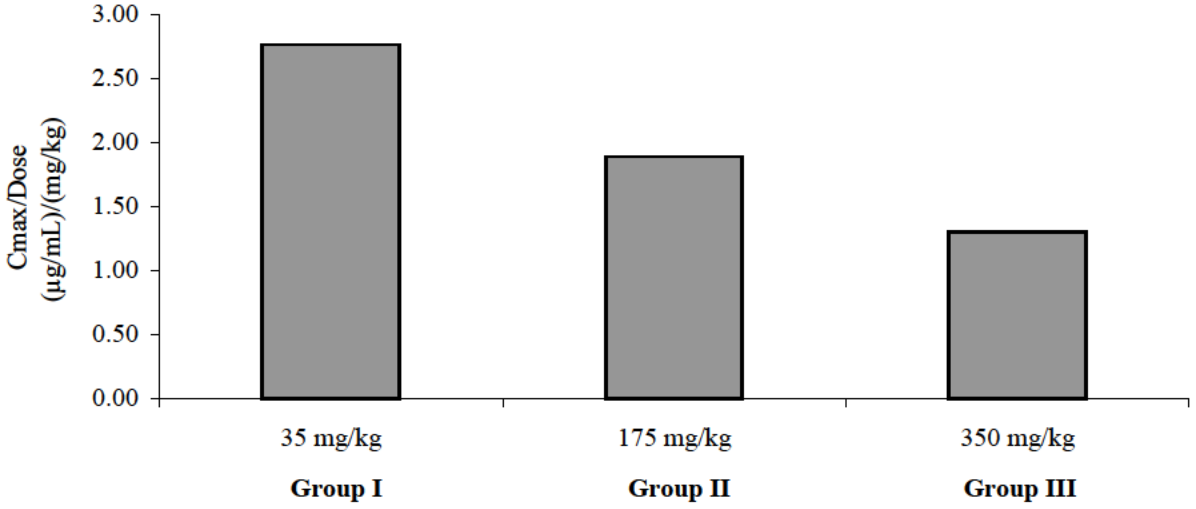


Figure 2 **Comparison of PFHxA Dose Normalized Exposure (Cmax/Dose and AUC(0-inf)/Dose) in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt**



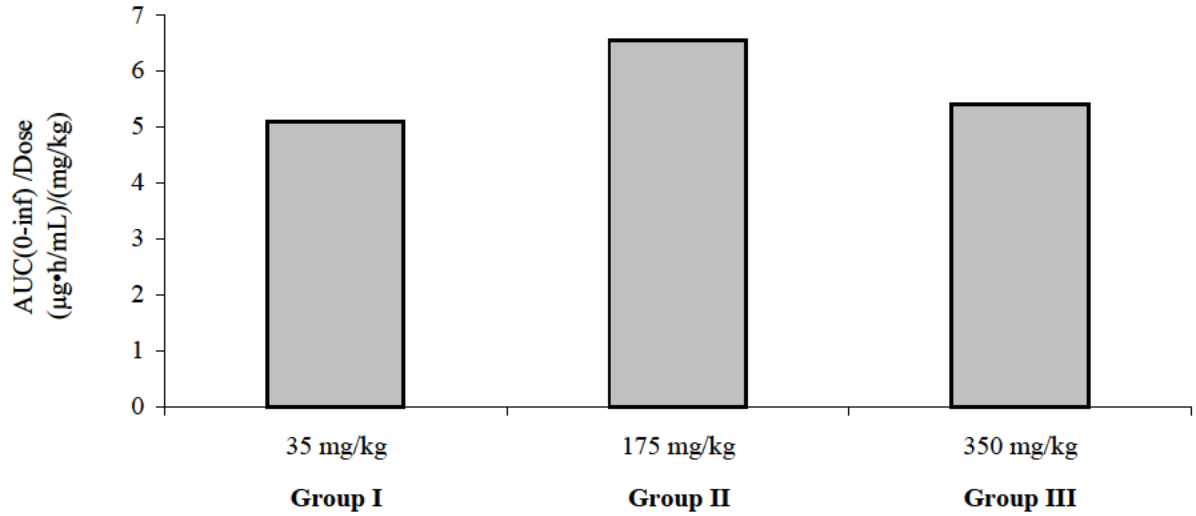
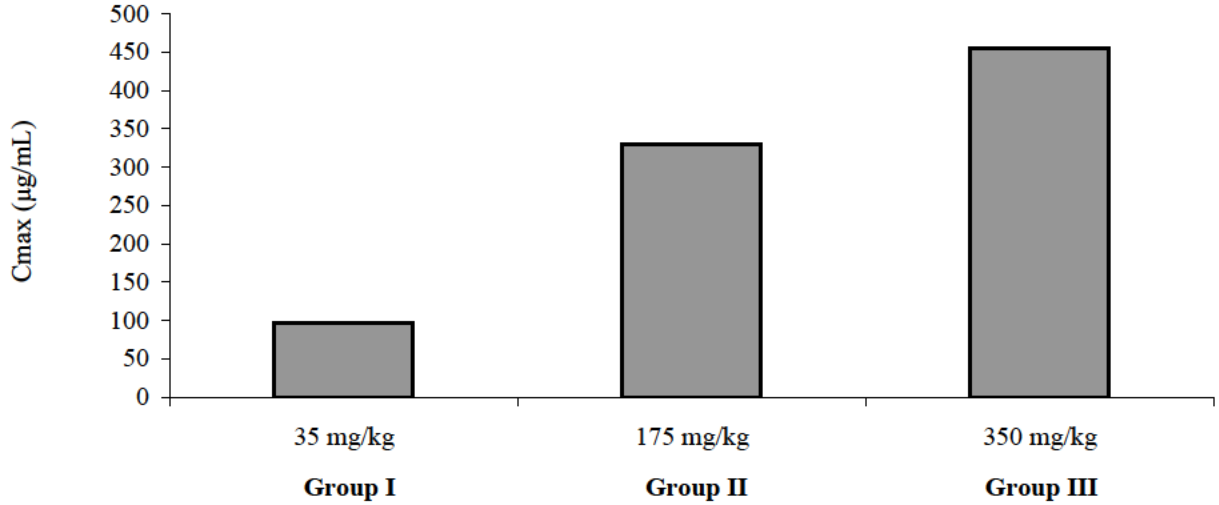
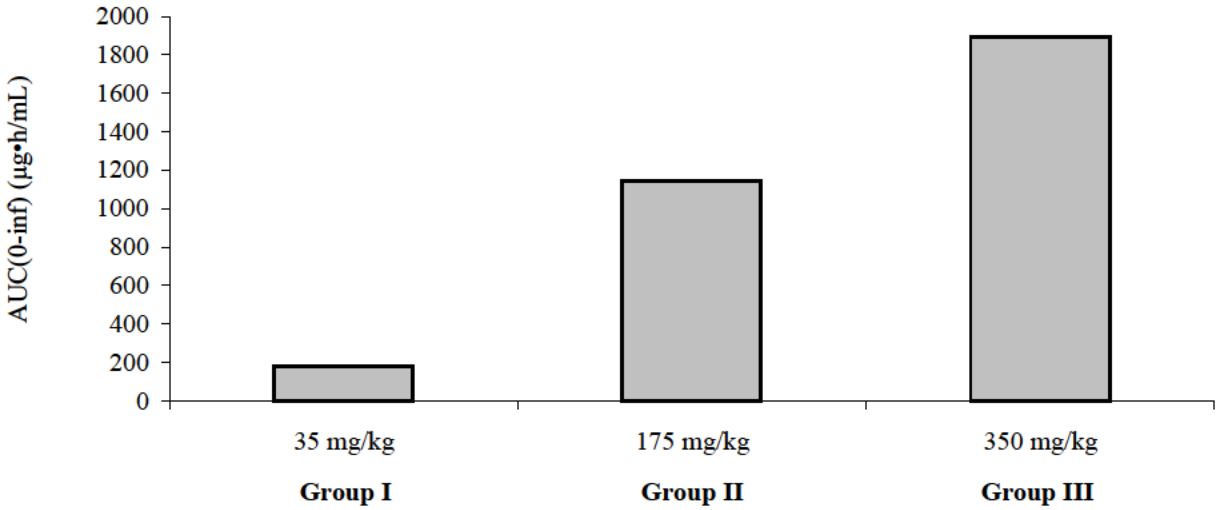


Figure 3 **Comparison of PFHxA Exposure (Cmax and AUC(0-inf)) in Female Crl:CD1(ICR) Mice Serum Following Oral Gavage of PFH Ammonium Salt**





Appendix 1 Data Capture/Entry Methods Used in Report

DATA CAPTURE/ENTRY METHODS USED IN REPORT

The following Data Capture/Entry methods have been used in this report:

Automated* data capture was performed by WinNonlin (version 3.2) from Pharsight Corporation, including values and units, which were then transferred into a Microcomputer using Excel (Microsoft Excel 2000/2003).

- Pharmacokinetics - Non-Compartmental Analysis

Data were transferred from an Excel environment into a Microcomputer Excel template (Microsoft Excel 2000/2003).

- Serum Concentration Analysis

Data transferred into Microcomputer template Excel (Microsoft Excel 2000/2003).

- Figures

* On occasion, retrospective manual entry may have been made for some value(s)/observation(s).

APPENDIX 5 - BIOANALYTICAL REPORT

FINAL REPORT

Test Site Ref. No. 141660
Testing Facility Study No. UZS00009

Determination of Perfluorohexanoic Acid (PFHxA) in Female Mouse Serum by Liquid Chromatography-tandem Mass Spectrometry (LC-MS/MS) in Support of Protocol No. UZS00009

TEST SITE:
Charles River Laboratories Preclinical Services Montreal
22022 Transcanadienne
Senneville, Quebec
Canada H9X 3R3

TESTING FACILITY:
Charles River Laboratories Preclinical Services
905 Sheehy Drive, Building A
Horsham, PA 19044
USA

SPONSOR:
Daikin Industries, Ltd.
Chemical Division
Umeda Center Building
4-12 Nakazaki-Nishi, 2-chrome
Kita-ku, Osaka 530-8323
Japan

12 April 2010

Page 1 of 34

TABLE OF CONTENTS

LIST OF TABLES 4

LIST OF FIGURES 5

LIST OF APPENDICES 6

1. COMPLIANCE STATEMENT 7

2. QUALITY ASSURANCE STATEMENT 8

3. SUMMARY 9

4. INTRODUCTION 10

5. REFERENCE STANDARD, INTERNAL STANDARD AND BLANK MATRIX 10
   - 5.1. Reference Standard 10
   - 5.2. Internal Standard 10
   - 5.3. Blank Matrix 10

6. EXPERIMENTAL PROCEDURES 11
   - 6.1. Calibration Standards 11
   - 6.2. Quality Control Samples 11
   - 6.3. Study Samples 11
   - 6.4. Incurred Sample Reanalysis 11
   - 6.5. Analysis 11
     - 6.5.1. Liquid Chromatography 12
     - 6.5.2. MS/MS Conditions 12
   - 6.6. System Suitability 13
   - 6.7. Data Collection and Statistical Methods 13
   - 6.8. Method Validation 13
   - 6.9. Quality Assurance 13
   - 6.10. Archives 13

7. PROTOCOL DEVIATIONS 14

8. RESULTS AND DISCUSSION 14
8.1. System Suitability 14
8.2. Study Samples 14
9. REPORT REVIEW AND APPROVAL SIGNATURE 15

LIST OF TABLES

Table 1 Group 1 Serum Concentrations of Perfluorohexanoic Acid 16

Table 2 Group 2 Serum Concentrations of Perfluorohexanoic Acid 17

Table 3 Group 3 Serum Concentrations of Perfluorohexanoic Acid 18

Table 4 Calibration Standard Statistics 19

Table 5 Quality Control Sample Statistics 20

Table 6 Incurred Sample Reanalysis 21

LIST OF FIGURES

Figure 1 Representative Calibration Line (Theoretical Concentration 1.00 to 1000 µg/mL) 22

Figure 2 Representative LLOQ Standard Chromatogram (Theoretical Concentration 1.00 µg/mL) 23

Figure 3 Representative ULOQ Standard Chromatogram (Theoretical Concentration 1000 µg/mL) 24

Figure 4 Representative Double Blank Chromatogram 25

Figure 5 Representative Sample Chromatogram (Group 1, Animal No. 3000, Day 1, Predose) 26

Figure 6 Representative Sample Chromatogram (Group 1, Animal No. 1204, Day 1, 0.5 h) 27

Figure 7 Representative Sample Chromatogram (Group 2, Animal No. 1222, Day 1, Predose) 28

Figure 8 Representative Sample Chromatogram (Group 2, Animal No. 1228, Day 1, 2 h) 29

Figure 9 Representative Sample Chromatogram (Group 3, Animal No. 1243, Day 1, Predose) 30

Figure 10 Representative Sample Chromatogram (Group 3, Animal No. 1252, Day 1, 4 h) 31

LIST OF APPENDICES

Appendix 1 Certificate of Analysis 32

I. COMPLIANCE STATEMENT [illegible]

This phase [illegible] Montreal (PCS-MTL) [illegible] with the Good [illegible] Administration (21 CFR [illegible] of the Organisation [illegible] work was not conducted [illegible] that the test site is [illegible] this agency.

[illegible] Date Date Date

[illegible] stigator
[illegible] lysis

Charles River [illegible]

2. QUALITY ASSURANCE STATEMENT

In compliance with [illegible] Good Laboratory Practice [illegible] Regulations [illegible] this study has been audited [illegible] the final report accurately [illegible] during the conduct of the study.

Phase	Date of Inspection	Dates of Reports to Study Director/ Principal Investigator	Dates of Reports to Testing Facility Management/ Study Director & Lead QA
Protocol Review	23 June 2009	25 June 2009	30 November 2009
Protocol Amendment Review	23 June 2009	25 June 2009	30 November 2009
Protocol Amendment Review	10 November 2009	[illegible] 2009	30 November 2009
SOP Review, Study-specific - In-life	09 November 2009	09 November 2009	30 November 2009
Bioanalytical Report Review	20 November 2009	25 November 2009	30 November 2009
Bioanalytical Data	25 November 2009		
Bioanalytical Report Review			
Bioanalytical Data	19 March 2010	29 March 2010	08 April 2010
Final Report Review	29 March 2010		
Sample Management/ Shipping Records Review			

In addition to the above mentioned inspections, [illegible] inspections were [illegible] of this study [illegible] these inspections [illegible] reports [illegible] Quality Assurance Standard Operating Procedures.

12 April 2010

Date

3. SUMMARY

The concentrations of perfluorohexanoic acid in female mouse serum samples in support of Testing Facility Study No. UZS00009, entitled "Oral (Gavage) Acute Pharmacokinetic Study of PFH Ammonium Salt (Ammonium Salt of Perflourinated Hexanoic Acid) in Mice," were determined using a previously validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) method. Results for all samples analyzed are presented in this report.

4. INTRODUCTION

The concentrations perfluorohexanoic acid in female mouse serum samples were determined by a liquid chromatography-tandem mass spectrometry (LC-MS/MS) method. The method of analysis, documented in PCS-MTL Analytical Procedure AP.141660.SE.02, was previously validated (Study No. 141837).

For the work detailed in this report, the experimental start date was 21 July 2009 and the experimental end date was 05 March 2010.

5. REFERENCE STANDARD, INTERNAL STANDARD AND BLANK MATRIX

5.1. Reference Standard

Identity:	PFH ammonium salt (50% aqueous solution: 474 mg/mL) (also known as perfluorohexanoic acid or PFHxA)
Lot number:	7005
Purity:	93.4% (correction factor: 0.474, corrected for effective component in solution)
Expiry date:	31 July 2010
Storage conditions:	Kept in a controlled temperature area set to maintain 21°C

5.2. Internal Standard

Identity:	Perfluoro-n-[1, 2-$^{13}C_2$] hexanoic acid (also known as PFHxA-1, 2-$^{13}C_2$)
Lot number:	MPFHxA0809
Purity:	> 98% (50 µg/mL certified solution)
Storage condition:	Kept in a refrigerator set to maintain 4°C, dark

The reference standard characterization was the responsibility of the Sponsor who provided a Certificate of Analysis (Appendix 1) for inclusion in this study report.

Details of identity, purity, storage conditions and handling precautions were supplied by the Sponsor. Remaining reference standard was stored at PCS-MTL for use on subsequent studies for the Sponsor.

5.3. Blank Matrix

Identity:	Female mouse serum
Species:	*Mus musculus*
Strain:	CD1

6. EXPERIMENTAL PROCEDURES

6.1. Calibration Standards

Calibration standards of reference standard were prepared in blank female mouse serum covering the theoretical concentration range of 1.00 to 1000 µg/mL. Calibration standards consisted of blank female mouse serum (250 µL) spiked with appropriate standard working solution (methanol; 5 µL).

6.2. Quality Control Samples

Quality control (QC) samples of reference standard were prepared in blank female mouse serum at theoretical concentrations of 3.00, 60.0 and 700 µg/mL. QC samples consisted of blank female mouse serum (250 µL) spiked with appropriate QC working solution (methanol; 5 µL).

6.3. Study Samples

Study samples were received from Charles River Laboratories Preclinical Services (Pennsylvania), and stored frozen in the freeze set to maintain at -80°C prior to analysis. Samples anticipated to be above the upper limit of quantitation (ULOQ) were diluted with blank female serum prior to initial analysis.

Remaining unused study samples will be retained at PCS-MTL for approximately 1 year after dispatch of the final report or until authorized to discard by the Study Director.

6.4. Incurred Sample Reanalysis

Incurred sample reanalysis (ISR) was conducted as per PCS-MTL SOP CAB-012.

6.5. Analysis

Single and double blank samples consisted of blank female mouse serum (250 µL) plus methanol (5 µL). To each standard, QC, single and double blank sample and study samples (10 µL), acetonitrile (100 µL) was added and the mixtures vortexed (~30 seconds) and centrifuged (~14000 rpm, ~10 minutes, set at 4°C). An aliquot (10 µL) of the supernatant was transferred to an appropriately labelled tube containing internal standard (100 ng/mL; 1.0 mL) or for double blank sample an aliquot (10 µL) of the supernatant was transferred to an appropriately labelled tube containing a solution of water:methanol (30:70, v/v; 1.0 mL) and the mixture vortexed. An aliquot (100 µL) of the mixture was transferred to a 96-well collection plate containing a solution of water:methanol (30:70, v/v; 900 µL) and the extracts vortexed (~30 seconds).

The standard, QC, blank and study sample extracts were analyzed by LC-MS/MS using the following conditions:

6.5.1. Liquid Chromatography

HPLC system: Agilent Technologies 1100 series binary pump and degasser, and Shimadzu SIL-HTC autosampler

Column: Waters XBridge Shield RP18, 3.5 µm (50 x 4.6 mm id)

Column temperature: Set at 50°C

Mobile phase gradient elution: Eluent A: 2 mM ammonium acetate, pH 4.0

Eluent B: methanol:2 mM ammonium acetate (pH 4.0); 80:20, v/v

Time (min)	%B
0.0	70
3.5	70

Flow rate: 1.0 mL/min

Injection volume: 5 µL

Autosampler tray temperature: Set at 4°C

Autosampler needle wash: Water:methanol:acetic acid; 20:80:1, v/v/v

6.5.2. MS/MS Conditions

MS system: MDS Sciex API 4000

Data capture system: MDS Sciex Analyst, Version 1.4.1

Ionization mode: Negative electrospray ionization (ESI)

Scan type: Multiple reaction monitoring (MRM)

Resolution: Unit/unit

Ion spray voltage: -4500 V

Ion source gas 1 (zero air): 60 psi

Ion source gas 2 (zero air): 60 psi

Curtain gas: 30 psi

Collision activated dissociation gas (CAD): 6 dacs

Temperature: 600°C

Monitoring ions and respective parameters:

Name	Q1 Mass	Q3 Mass	Retention Time (min)	Scan Time (msec)	DP (V)	EP (V)	CE (eV)	CXP (V)
Perfluoro-n-hexanoic acid	313.0	268.8	~2.5	200	-40	-5	-13	-15
PFHxA-^{13}C2	315.0	270.0	~2.5	100	-40	-5	-13	-15

Some conditions may vary.

6.6. System Suitability

The reproducibility of the chromatographic system was determined by injecting an extracted calibration standard, at least in triplicate, at the beginning of the chromatographic run. To assess system stability, QC samples were injected at the end of each run.

A coefficient of variation (CV) of ≤ 5% with respect to peak area ratio for an extracted calibration standard injected at the beginning of the run, and QC samples injected at the end of each run meeting acceptance criteria, were considered acceptable.

6.7. Data Collection and Statistical Methods

Data collection was performed using Analyst, Version 1.4.1, from MDS Sciex.

Statistical analyses included quadratic regression with 1/concentration^2 weighting and descriptive statistics such as arithmetic means and standard deviations, accuracy and precision using Watson Laboratory Information Management System (LIMS) (Version 7.2.0.02) and Microsoft Excel (Version 2000/2003).

Tables were prepared from retrospective manual entry on computer (Microsoft Word, Version 2000/2003).

6.8. Method Validation

The analytical method was previously validated (Study No. 141837) with respect to selectivity, linearity, lower limit of quantitation (LLOQ), carry-over, intra- and inter-assay precision and accuracy, stock solution stability, injection medium integrity, short-term matrix stability, freeze-thaw matrix stability, long-term matrix stability and dilution integrity.

6.9. Quality Assurance

The Quality Assurance department of PCS-MTL undertook and documented inspections and process audits of the laboratories in which this study was performed at PCS-MTL, and audited the study report as well as the raw data. The Quality Assurance Statement is presented on Page 8.

6.10. Archives

Samples, reference standard, all raw data and documents generated at PCS-MTL during this study, together with the final report will be retained in the scientific archives of PCS-MTL for approximately one year after dispatch of the final report. Subsequent storage details will be documented in the raw data.

7. PROTOCOL DEVIATIONS

As per study protocol, the study was to comply with regulations from the Ministry of Agriculture, Forestry and Fisheries; however, the bioanalytical portion of the study did not comply with this. This deviation is considered to have no impact on the outcome of bioanalytical portion of the study as the work conducted was compliant to the other regulation agencies mentioned above, and that the work was reviewed by the Quality Assurance Unit and that all results were within acceptance criteria

8. RESULTS AND DISCUSSION

A representative calibration line is presented in Figure 1, and representative chromatograms are presented in Figure 2, Figure 3, Figure 4, Figure 5, Figure 6, Figure 7, Figure 8, Figure 9 and Figure 10.

8.1. System Suitability

Acceptance criteria with respect to system suitability were met on all occasions.

8.2. Study Samples

All study samples were previously analyzed using a validated analytical method having an analytical range of 0.300 to 200 ng/mL (validation Study No. 141658). However, serum concentration of the samples was observed to be very high in the initial analysis. In addition, significant carry-over from high concentration study samples was observed and the analytical runs (runs 01 and 02) were rejected for this reason. Based on the observed concentration, it was judged that the analytical range in the initial method was inadequate for the analysis of the study samples and thus the method was re-validated (validation Study No. 141837) with a higher analytical range (1.00 to 1000 µg/mL). All study samples were re-analyzed with the new method. Data analyzed with the initial method is not reported but is kept on file with the raw data study binder.

Results for the study samples are presented in Table 1, Table 2 and Table 3. The calibration standard and quality control sample statistics are presented in Table 4 and Table 5, respectively.

Incurred sample re-analysis was successfully performed as per PCS-MTL SOP CAB-012. The results are presented in Table 6.

9. REPORT TESTING FACILITY AND SPONSOR SIGNATURE

This report has been reviewed by and approved by:

12th February 2010

Date Date Date

Table 1 Group 1 Serum Concentrations of Perfluorohexanoic Acid

Animal	Dosage Group	Study Day	Nominal Time (hour)	Concentration (µg/mL)
3000	1	1	0	< LLOQ
3001	1	1	0	< LLOQ
3002	1	1	0	< LLOQ
1204	1	1	0.5	132
1205	1	1	0.5	65.3
1206	1	1	0.5	92.8
1207	1	1	2	36.0
1208	1	1	2	22.4
1209	1	1	2	49.7
1210	1	1	4	6.73
1211	1	1	4	12.1
1212	1	1	4	3.90
1213	1	1	6	3.25
1214	1	1	6	< LLOQ
1215	1	1	6	1.53
1216	1	1	8	< LLOQ
1217	1	1	8	< LLOQ
1218	1	1	8	< LLOQ
1219	1	1	24	< LLOQ
1220	1	1	24	< LLOQ
1221	1	1	24	< LLOQ

LLOQ = lower limit of quantiation (theoretical concentration 1.00 µg/mL)

Table 2 Group 2 Serum Concentrations of Perfluorohexanoic Acid

Animal	Dosage Group	Study Day	Nominal Time (hour)	Concentration (µg/mL)
1222	2	1	0	< LLOQ
1223	2	1	0	< LLOQ
1224	2	1	0	< LLOQ
1225	2	1	0.5	364
1226	2	1	0.5	288
1227	2	1	0.5	338
1228	2	1	2	317
1229	2	1	2	360
1230	2	1	2	264
1231	2	1	4	69.9
1232	2	1	4	118
1233	2	1	4	101
1234	2	1	6	39.5
1235	2	1	6	8.37
1236	2	1	6	8.31
1237	2	1	8	2.11
1238	2	1	8	2.38
1239	2	1	8	34.4
1240	2	1	24	< LLOQ
1241	2	1	24	2.02
1242	2	1	24	< LLOQ

LLOQ = lower limit of quantiation (theoretical concentration 1.00 µg/mL)

Table 3 Group 3 Serum Concentrations of Perfluorohexanoic Acid

Animal	Dosage Group	Study Day	Nominal Time (hour)	Concentration (µg/mL)
1243	3	1	0	< LLOQ
1244	3	1	0	< LLOQ
1245	3	1	0	< LLOQ
1246	3	1	0.5	381
1247	3	1	0.5	509
1248	3	1	0.5	473
1249	3	1	2	440
1250	3	1	2	124
1251	3	1	2	301
1252	3	1	4	317
1253	3	1	4	332
1254	3	1	4	229
1255	3	1	6	199
1256	3	1	6	103
1257	3	1	6	168
1258	3	1	8	24.1
1259	3	1	8	12.2
1260	3	1	8	7.45
1261	3	1	24	2.68
1262	3	1	24	< LLOQ
1263	3	1	24	< LLOQ

LLOQ = lower limit of quantiation (theoretical concentration 1.00 µg/mL)

Table 4 Calibration Standard Statistics

Analytical Run[a]	Concentration (µg/mL)									
	1.00	2.00	5.00	25.0	50.0	100	200	400	800	1000
3	0.992	2.02	5.08	24.2	50.2	101	200	401	800	999
4	1.00	2.01	4.85	24.9	50.2	102	203	414	680	1109
5	0.970	2.13	4.93	25.0	49.0	102	194	405	763	1038
Mean	0.9884	2.056	4.952	24.67	49.80	101.7	199.0	406.9	747.4	1048.6
S.D.	0.01647	0.0653	0.1159	0.448	0.717	0.83	4.50	6.78	61.30	56.03
%CV	1.7	3.2	2.3	1.8	1.4	0.8	2.3	1.7	8.2	5.3
% Bias	-1.2	2.8	-1.0	-1.3	-0.4	1.7	-0.5	1.7	-6.6	4.9
n	3	3	3	3	3	3	3	3	3	3

a = Runs 01 and 02 were analyzed with initial analytical method with a lower analytical range; runs were also rejected due to significant carry-over from high concentration study samples. Results are not included in statistical calculation. Runs 06 and 07 were used for the assessment of long-term matrix stability which will be reported under validation Study No. 141837, no study samples were analyzed, thus the runs are not included in the statistical calculation.

Table 5 Quality Control Sample Statistics

Analytical Run[a]	Concentration (µg/mL)		
	3.00	60.0	700
3	3.15	64.5	693
	3.32	65.6	684
	3.44	64.9	681
	3.33	64.1	677
4	3.34	63.1	741
	3.20	60.2	725
	3.26	64.5	787
	3.25	63.5	778
5	3.25	63.2	665
	3.11	61.2	699
	3.39	62.1	760
	3.21	59.1	728
Mean	3.271	63.00	718.1
S.D.	0.0970	2.000	41.50
%CV	3.0	3.2	5.8
% Bias	9.0	5.0	2.6
n	12	12	12

a = Runs 01 and 02 were analyzed with initial analytical method with a lower analytical range; runs were also rejected due to significant carry-over from high concentration study samples. Results are not included in statistical calculation. Results are not included in statistical calculation. Runs 06 and 07 were used for the assessment of long-term matrix stability which will be reported under validation Study No. 141837, no study samples were analyzed, thus the runs are not included in the statistical calculation.

Table 6 Incurred Sample Reanalysis

Animal	Dosage Group	Study Day	Nominal Time (hour)	Original Conc. (µg/mL)	Reassay Conc. (µg/mL)	% Difference
1204	1	1	0.5	132	124	-6.1
1205	1	1	0.5	65.3	66.4	1.6
1207	1	1	2	36.0	31.8	-11.7
1208	1	1	2	22.4	21.3	-5.0
1210	1	1	4	6.73	6.10	-9.3
1211	1	1	4	12.1	11.2	-7.6
1225	2	1	0.5	364	343	-5.9
1226	2	1	0.5	288	378	31.3
1228	2	1	2	317	330	4.2
1229	2	1	2	360	360	0.1
1231	2	1	4	69.9	63.7	-8.9
1232	2	1	4	118	127	7.3
1234	2	1	6	39.5	37.7	-4.6
1235	2	1	6	8.37	8.42	0.6
1247	3	1	0.5	509	494	-2.9
1251	3	1	2	301	417	38.6
1253	3	1	4	332	325	-2.3
1255	3	1	6	199	193	-2.9
1256	3	1	6	103	106	2.3
1258	3	1	8	24.1	25.0	3.7

% difference = {(reassay conc. - original conc.)/original conc.} X 100

Figure 1 **Representative Calibration Line (Theoretical Concentration 1.00 to 1000 µg/mL)**

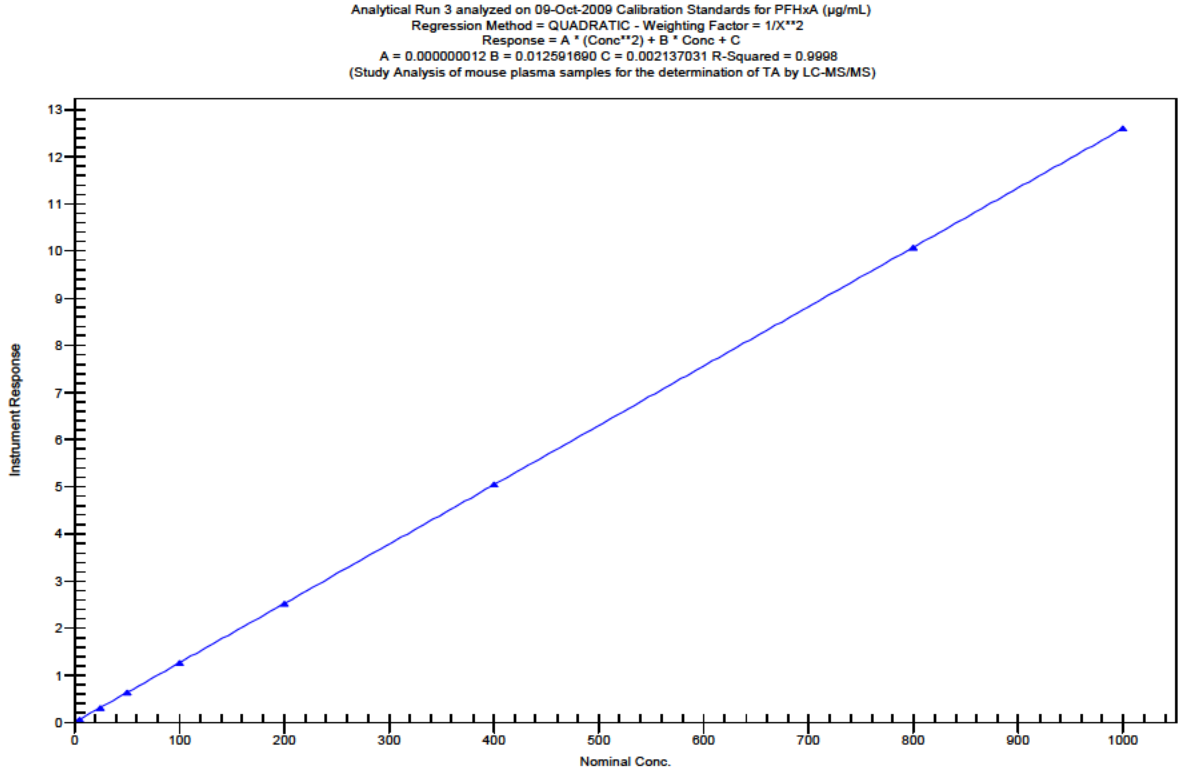


Figure 2 **Representative LLOQ Standard Chromatogram (Theoretical Concentration 1.00 µg/mL)**

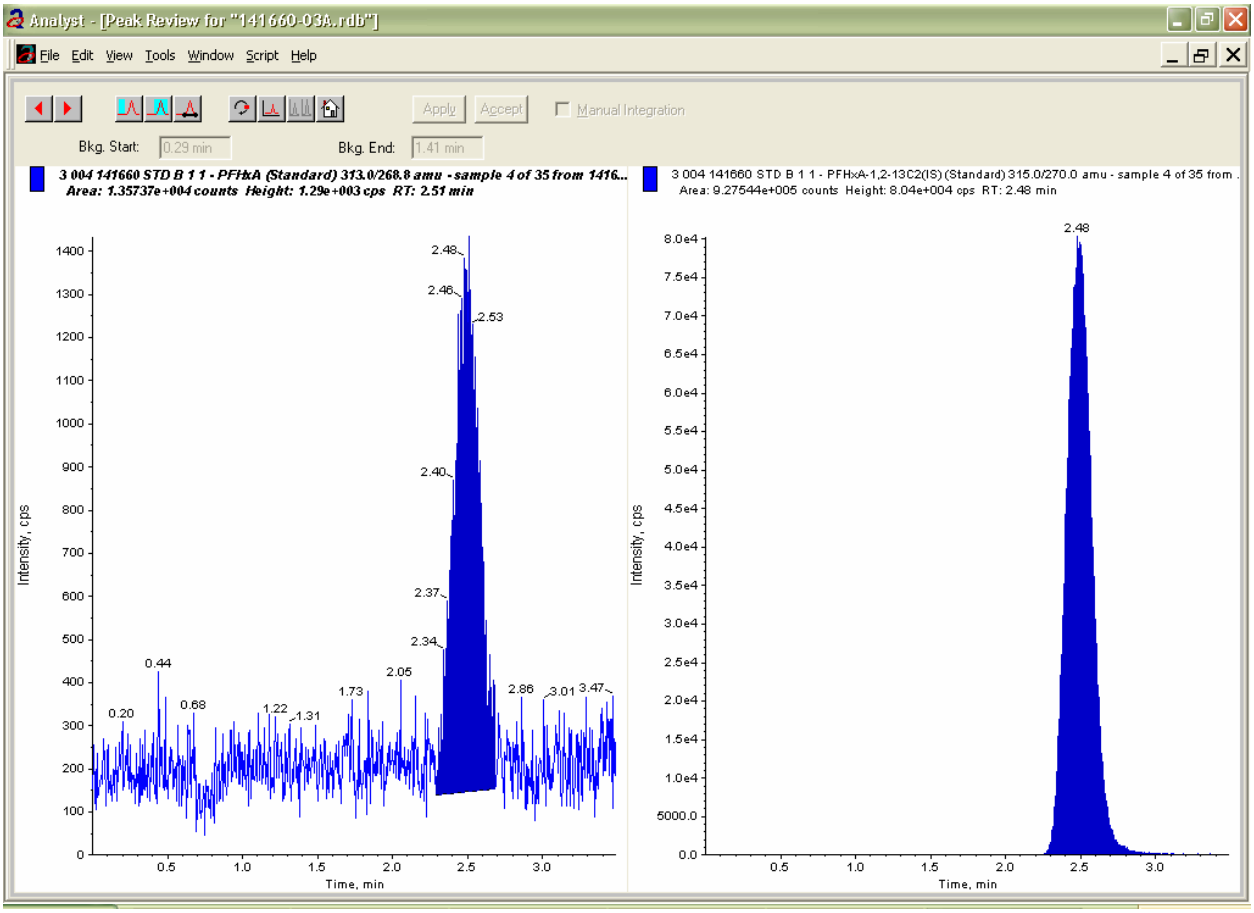


Figure 3 **Representative ULOQ Standard Chromatogram (Theoretical Concentration 1000 µg/mL)**

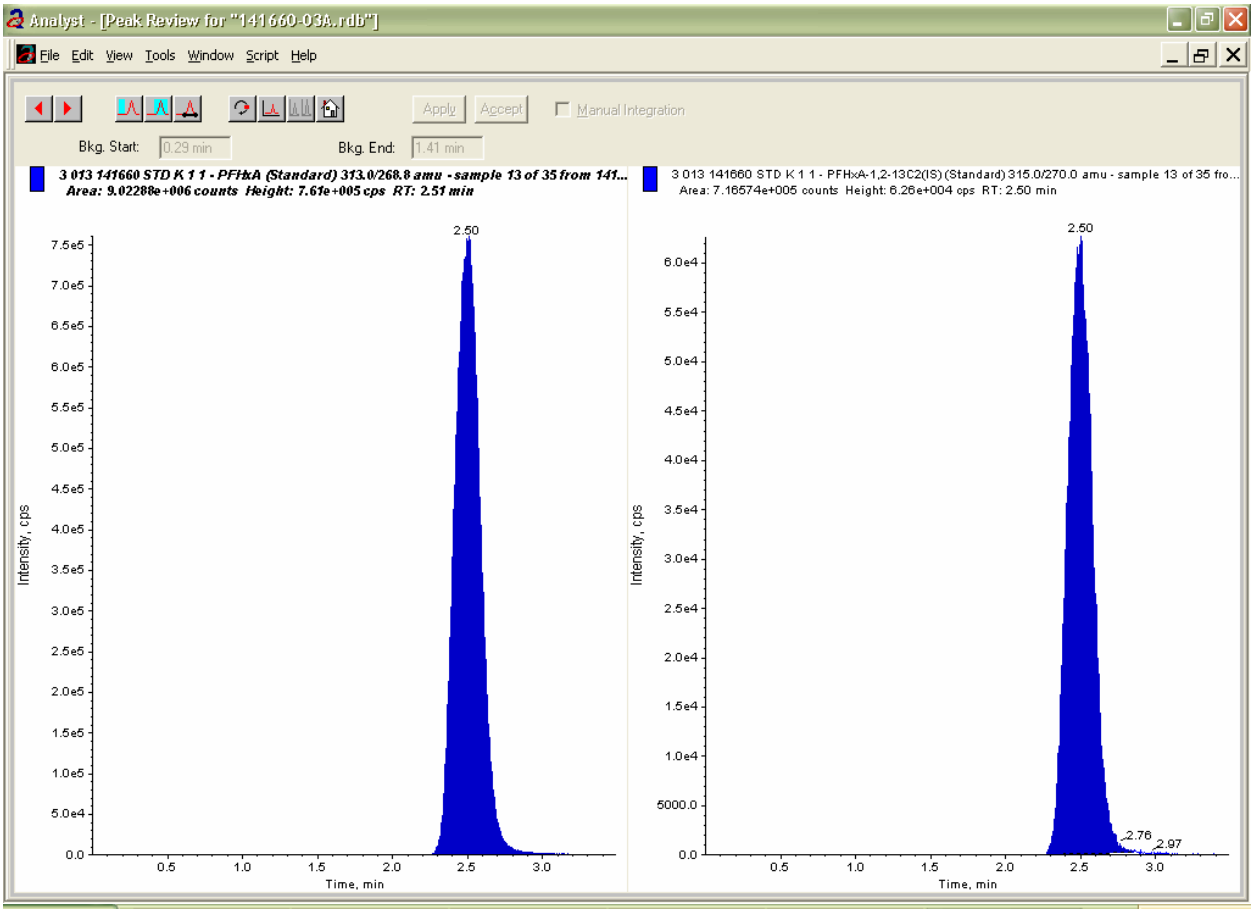


Figure 4 Representative Double Blank Chromatogram

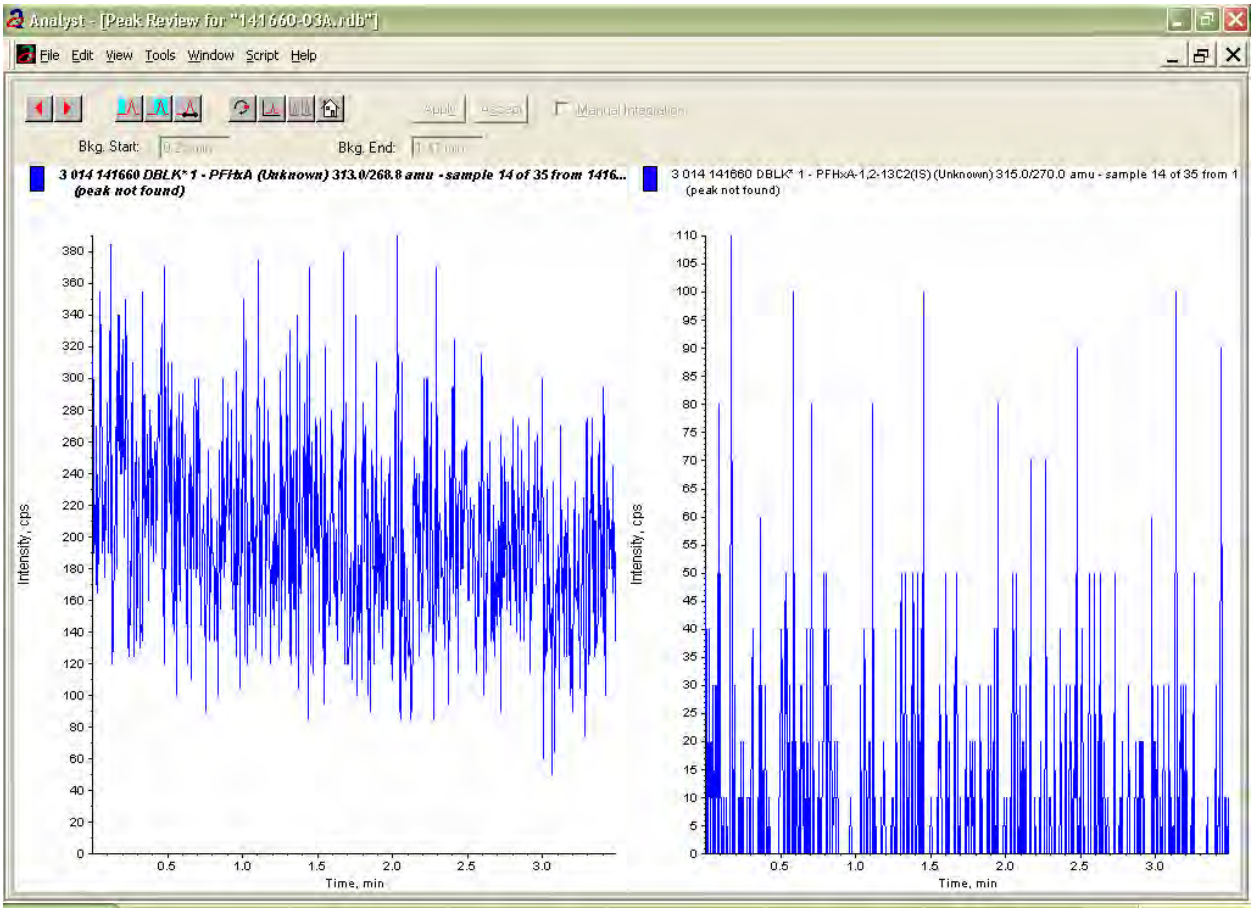


Figure 5 **Representative Sample Chromatogram (Group 1, Animal No. 3000, Day 1, Predose)**

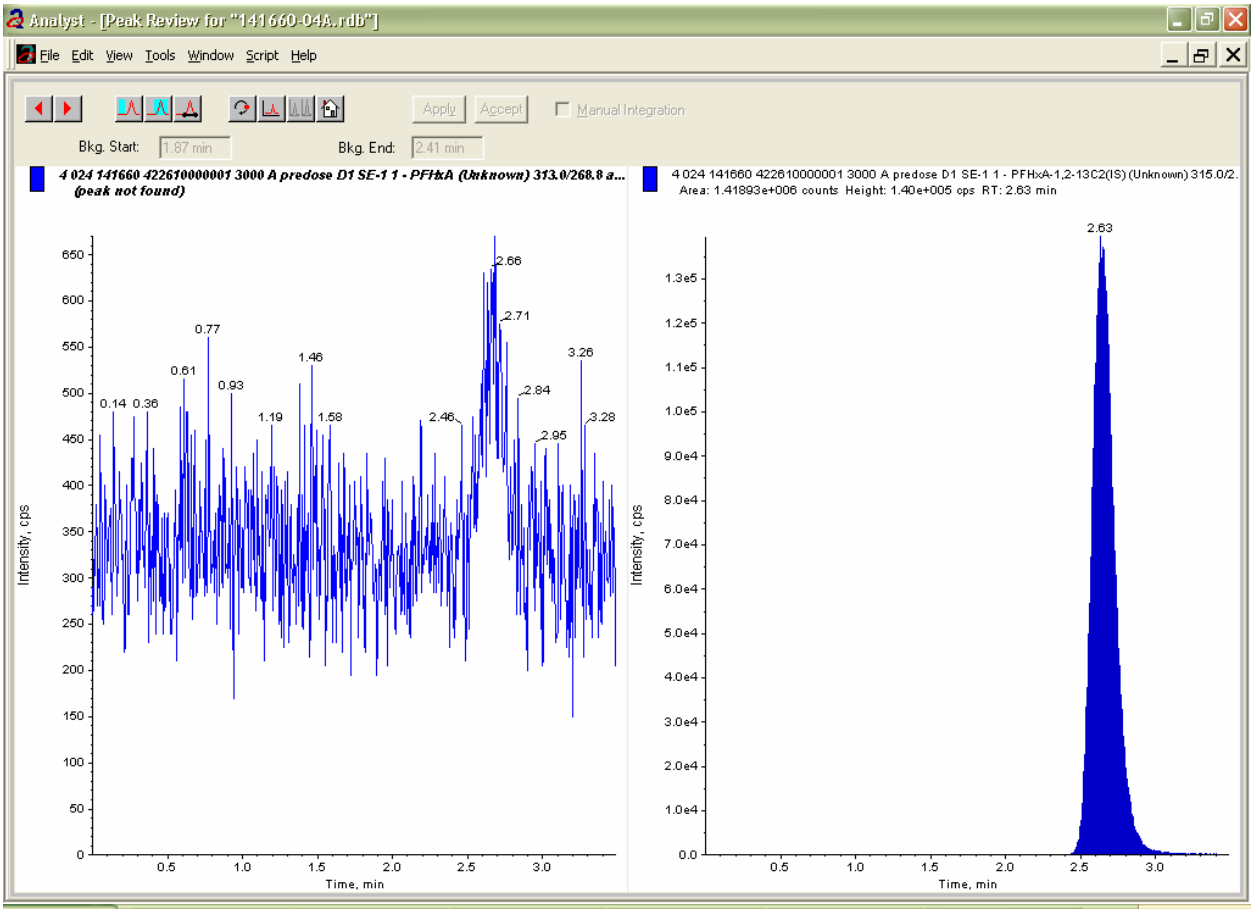


Figure 6 **Representative Sample Chromatogram (Group 1, Animal No. 1204, Day 1, 0.5 h)**

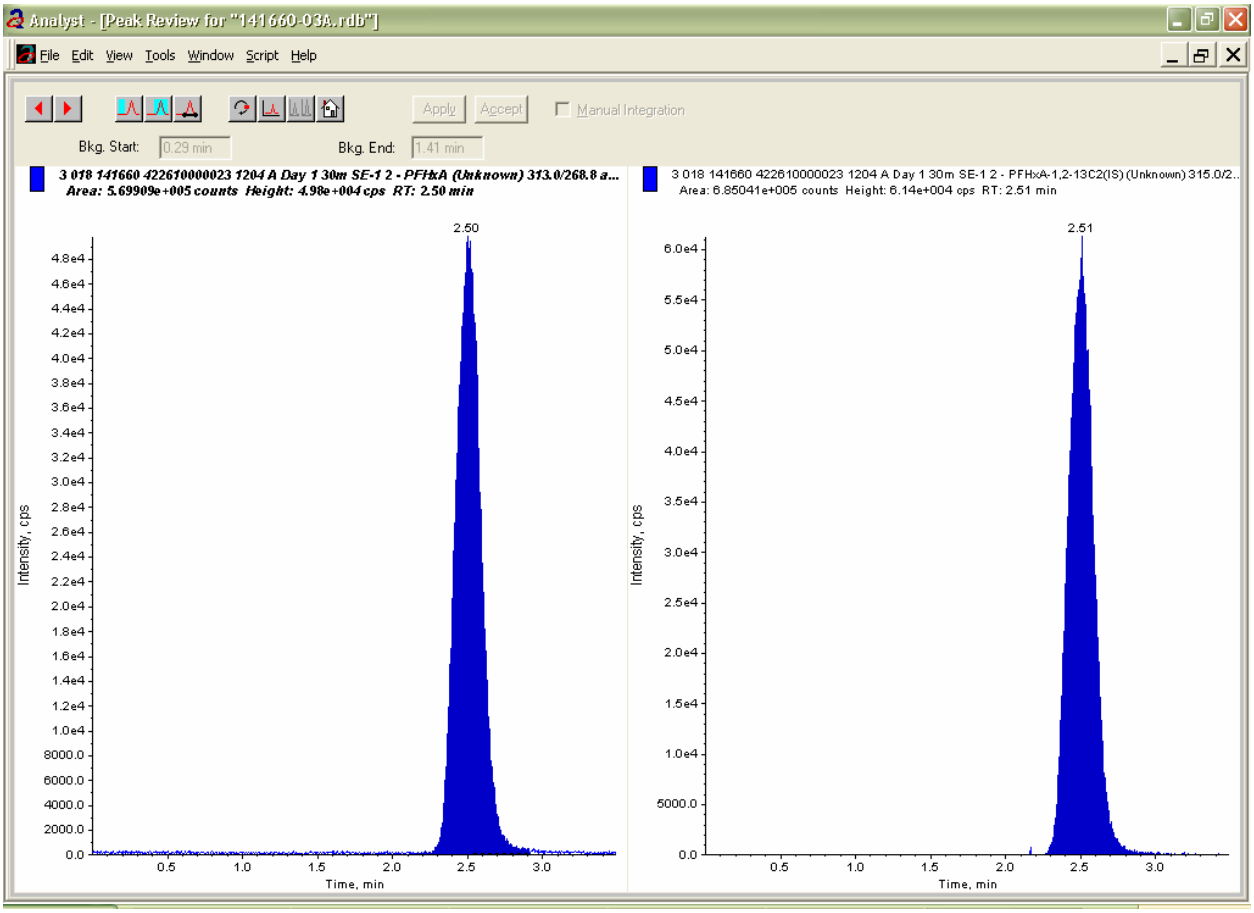


Figure 7 **Representative Sample Chromatogram (Group 2, Animal No. 1222, Day 1, Predose)**

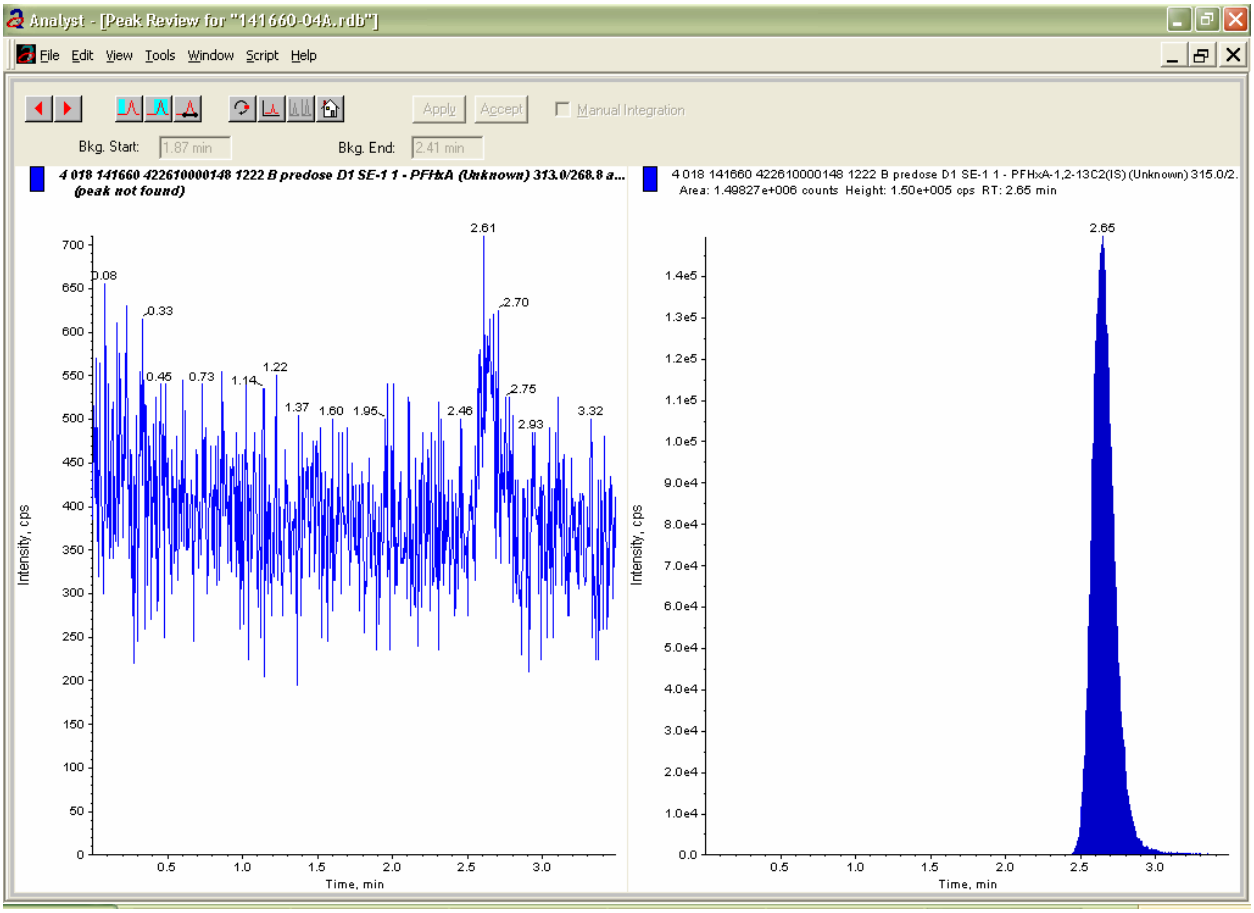


Figure 8 **Representative Sample Chromatogram (Group 2, Animal No. 1228, Day 1, 2 h)**

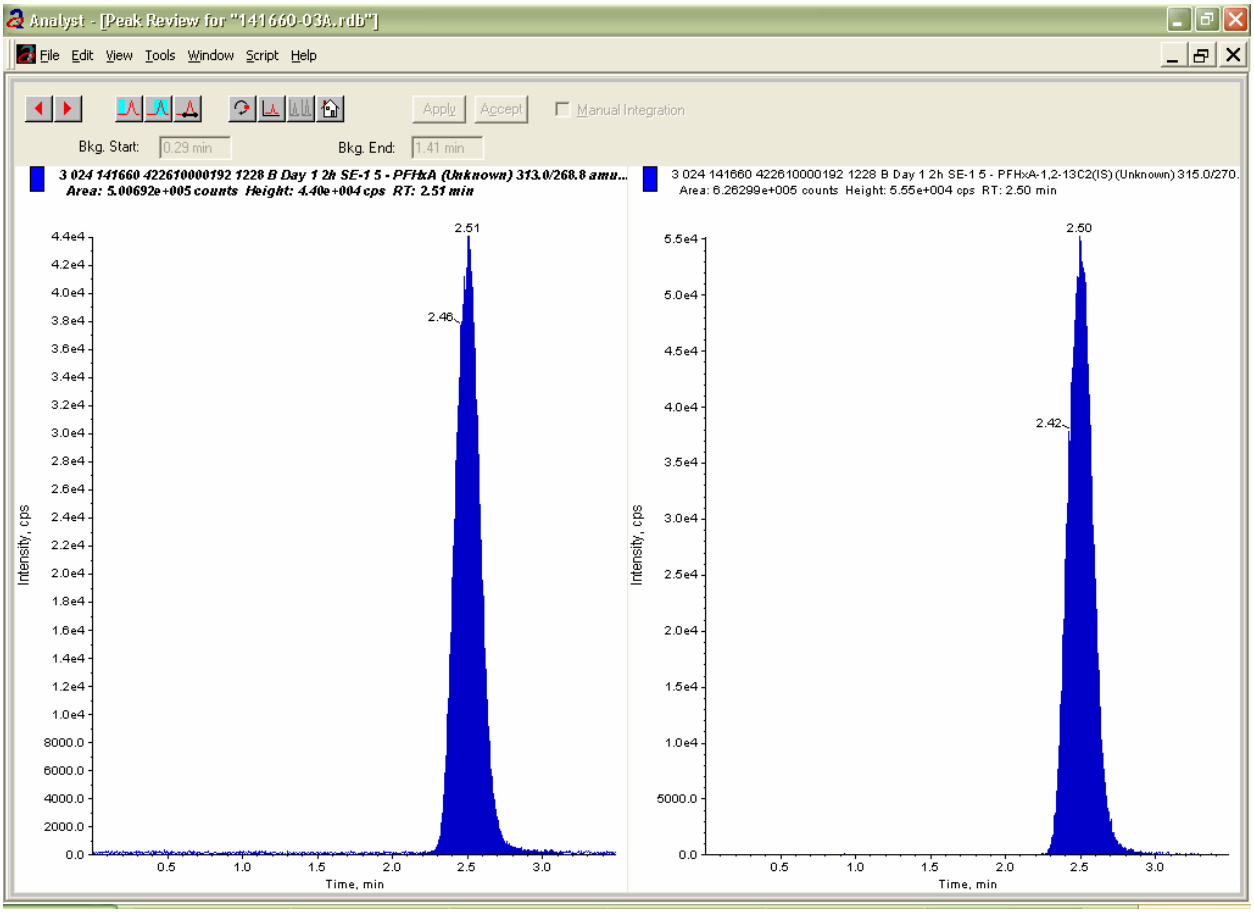


Figure 9 **Representative Sample Chromatogram (Group 3, Animal No. 1243, Day 1, Predose)**

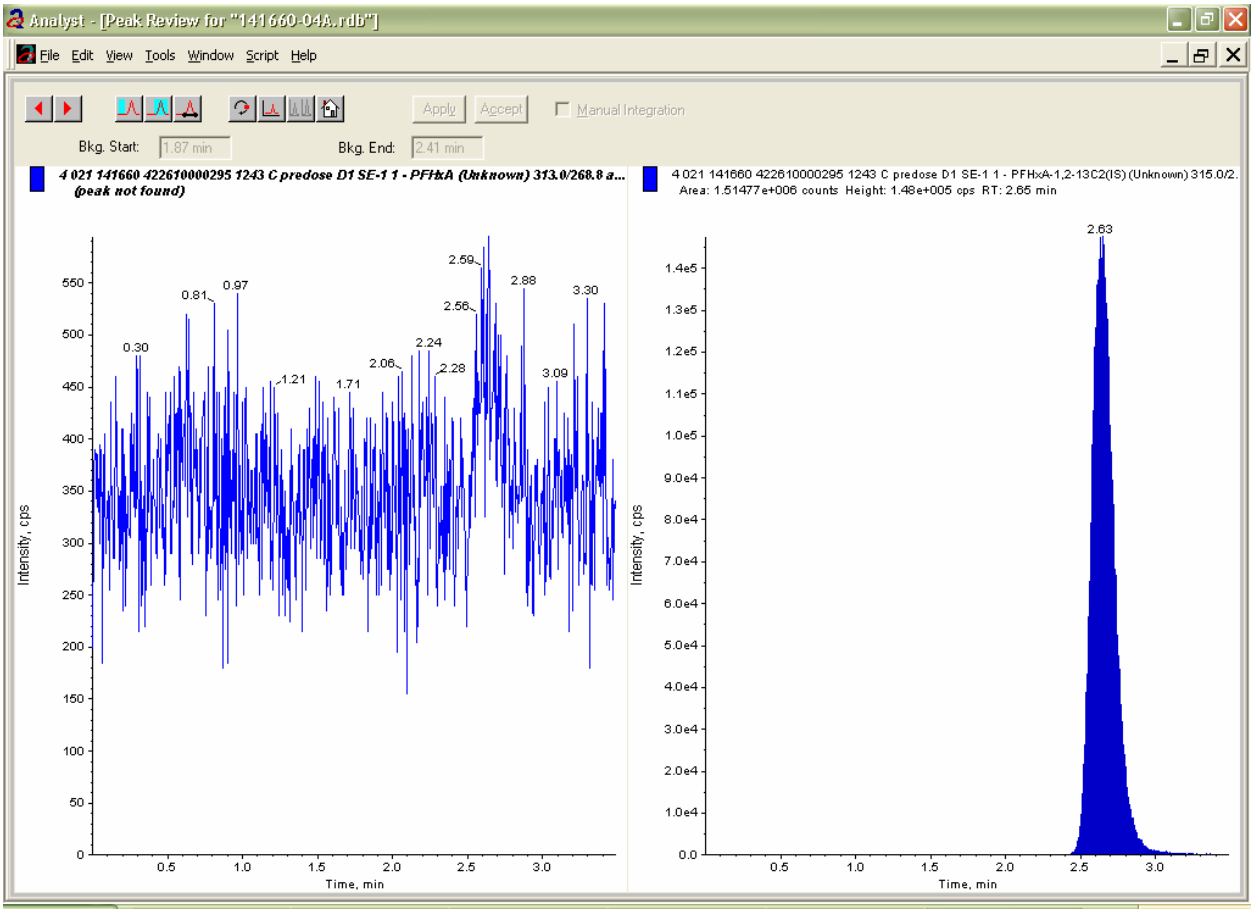
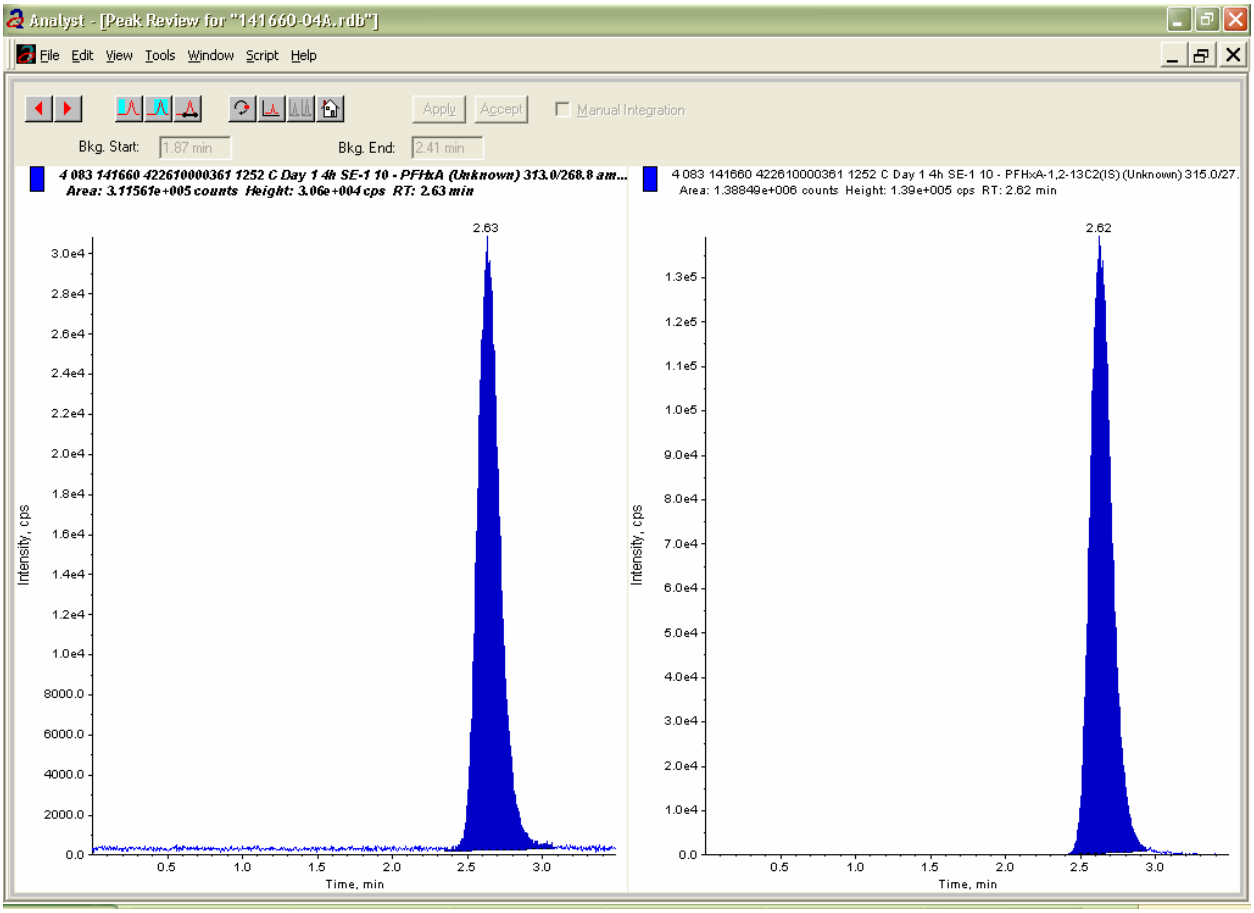


Figure 10 **Representative Sample Chromatogram (Group 3, Animal No. 1252, Day 1, 4 h)**



Appendix 1 Certificate of Analysis

Certificate of Analysis

Daikin Industries,LTD.

Name of Sample	*PFH Ammonium Salt（C-1500N）*
Lot.	*7005*
Date of Analysis	*May 14, 2009*
Purify	*47.4% (Effective component in Water)*
	**50.8*0.934%=47.4%*

COMPOSITION

identity		*Conc.*
#1	*Ammonium Perfluorohexanoate CAS RN. 21615-47-4*	*93.4%*
#2	*Unknown*	*6.6%*
	Total	*100%*

Analysis system (HPLC)

Equipment	*: Waters Alliance2695*
Detector	*: Waters 2487UV*
Detection wavelength	*: 210nm*

Analysis condition

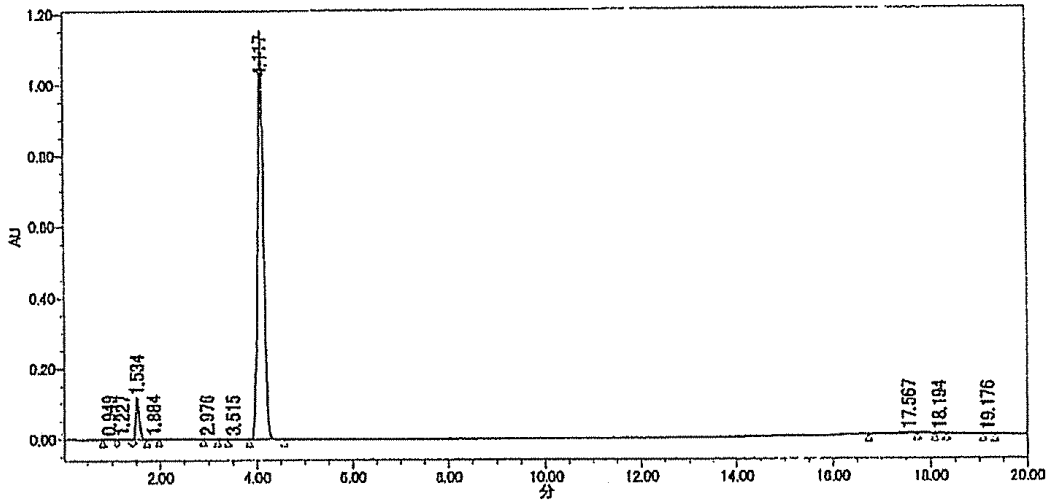
Column	*: TOSOH TSKGel ODS120T 4.6mm×150mm*
Temp.	*: 40 ℃*
Mobile phase	*: A=acetonitrile 、B=Solution of 0.6% perchloric acid in water*
Gradient	*: A:B=50:50(mass%) (0-10min.) →90:10(mass%) (15-20min.)*
Injection volume	*: 20μL*
Injection Concentration	*: 1%（dilute 50times with water)*

Chemical R&D Center
Unidyne Group
Senior Researcher

SIGNATURE
DATE : May 18, 2009

Analysis

サンプル名:	C1500N	分析担当者:	System
サンプルの種類:	未知試料	分析日:	2009/05/14 11:49:44
バイアル:	82	取り込みメソッドセット:	090514S
注入#:	1	解析日:	2009/05/14 13:55:17
注入量:	20.00 μl	解析メソッド:	C1500N
分析時間:	20.00 分	チャンネル名:	2487チャンネル 1
サンプルセット名:		解析チャンネルの説明:	



	成分名	Retention Time(min.)	Area (μVsec.)	Area (%)	Height (μV)		
1		0.949	17634	0.18	2554		
2		1.227	20551	0.20	1927		
3		1.534	574660	5.71	116134		
4		1.884	5543	0.06	710		
5		2.976	2424	0.02	414		
6		3.515	4940	0.05	361		
7		4.117	9390042	93.38	1144218		
8		17.567	29475	0.29	984		
9		18.194	6956	0.07	1098		
10		19.176	3881	0.04	592		

1/1 ページ

APPENDIX 6 - ENVIRONMENTAL AND HUSBANDRY REPORTS

TEMPERATURE AND RELATIVE HUMIDITY

ARGUS

Temperature and Relative Humidity Report
Location: Room 05

Protocol Number: UZS00009

Range of Dates: 19-May-2009 13:18 to 28-May-2009 15:59

	Temperature		Relative Humidity	
Target Range:	64°F to 79°F		30% to 70%	
Species: Mouse				
Total Number of Days:	10		10	
Total Number of Hours:	218.25		218.25	
Total Number of Data Points:	219		219	
Mean (± SD):	71.9	(± 0.5)	55.7	(± 5.6)
Maximum:	73.2		66.5	
Median:	71.9		55.5	
Minimum:	70.5		44.7	
Number of Points in Range (%):	219	(100.0)	219	(100.0)
Number of Points High (%):	0	(0.0)	0	(0.0)
Number of Points Low (%):	0	(0.0)	0	(0.0)

Report Generated: 04-Jun-2009 at 08:48

COMMENTS: _____

REVIEWED BY: _____ **DATE:** 04JUN09

ARGUS

Temperature and Relative Humidity Report
Location: Room 16

Protocol Number: UZS00009

Range of Dates: 10-Nov-2009 10:08 to 12-Nov-2009 14:59

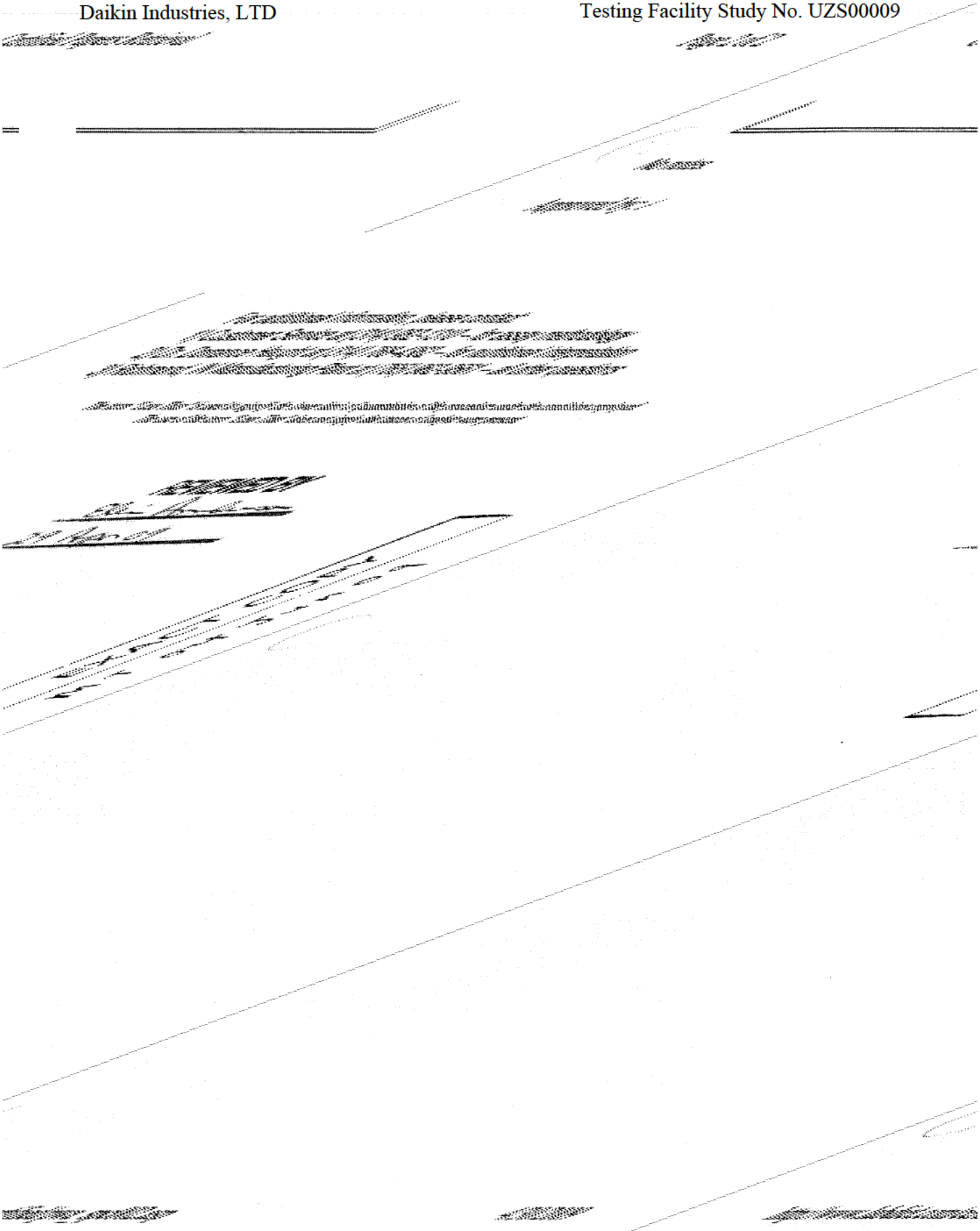
	Temperature		Relative Humidity	
Target Range:	64°F to 79°F		30% to 70%	
Species: Rat				
Total Number of Days:	3		3	
Total Number of Hours:	52.5		52.5	
Total Number of Data Points:	53		53	
Mean (± SD):	70.9	(± 1.1)	47.4	(± 3.3)
Maximum:	73.7		55.0	
Median:	71.0		46.7	
Minimum:	68.9		41.5	
Number of Points in Range (%):	53	(100.0)	53	(100.0)
Number of Points High (%):	0	(0.0)	0	(0.0)
Number of Points Low (%):	0	(0.0)	0	(0.0)

Report Generated: 18-Nov-2009 at 14:29

COMMENTS: _____

REVIEWED BY: _____ **DATE:** 18 Nov 2009

FEED ANALYSIS



LabDiet PMI NUTRITION

Return to Certified Analysis Retrieval

Product Code: 5002
Product Desc: CERTIFIED RODENT DIET
Lab Number: L0921776-2
Lot Code: SEP 02 09 3B
Entered: 9/21/2009

Assay	Analysis	Units
PROTEIN	20.8	%
FAT (ACID HYDRO)	5.6	%
FIBER (CRUDE)	5.07	%
ARSENIC	LESS THAN 0.2	PPM
CADMIUM	0.0911	PPM
CALCIUM	0.8132	%
LEAD	0.199	PPM
MERCURY	LESS THAN 0.025	PPM
PHOSPHORUS	0.6357	%
SELENIUM	0.415	PPM

Organophosphates	PPM	Organophosphates	PPM
Diazinon	LESS THAN 0.02	Disulfoton	LESS THAN 0.02
Ethion	LESS THAN 0.02	Malathion	LESS THAN 0.02
Methyl Parathion	LESS THAN 0.02	Parathion	LESS THAN 0.02
Thimet	LESS THAN 0.02	Trithion	LESS THAN 0.02

Chlorinated Hydrocarbons and PCB	PPM	Chlorinated Hydrocarbons and PCB	PPM
Aldrin	LESS THAN 0.02	Alpha-BHC	LESS THAN 0.02
Beta-BHC	LESS THAN 0.02	Chlordane	LESS THAN 0.02
DDE	LESS THAN 0.02	DDT	LESS THAN 0.02
Delta-BHC	LESS THAN 0.02	Dieldrin	LESS THAN 0.02
Endrin	LESS THAN 0.02	HCB	LESS THAN 0.02
Heptachlor	LESS THAN 0.02	Heptachlor Epoxide	LESS THAN 0.02
Lindane	LESS THAN 0.02	Methoxychlor	LESS THAN 0.02
Mirex	LESS THAN 0.02	PCB	LESS THAN 0.15
Thiodan	LESS THAN 0.02		

AFLATOXINS	PPB Aflatoxins	LESS THAN 5

EXACT COPY
HDV 22 Apr 2010

Approved
08 OCT 09

No notes.

Approved
06 OCT 2009

For additional information, please contact:
1) Customer Service at (314) 982-1310 – for assay methodology
2) Dr. Kristi Thompson, (765)894-3104 or Dr. Carrie Schultz, (314)974-6529 – for nutritional interpretation
3) Richmond, IN Manufacturing Plant at (765) 962-9561 -- all other questions

The term "Less Than" is used to signify the lower limit of quantitation of the procedure under the conditions employed.
The use of the term "Less Than" does not imply that traces of analyte were present.

EXACT COPY
MPV 22 APR 2010

WATER ANALYSIS

QC Laboratories

Analytical Report

ACCREDITED IN ACCORDANCE WITH nelac
05/05/09 02:23pm

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Regarding:

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1088907

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-1	DRINKING WATER - INVITRO LAB Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:03am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL LOW LEVEL-FIELD	SM 4500CL G	< 0.02 mg/l		05/01/09 11:03AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:42PM DH2
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:42PM DH2

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-2	DRINKING WATER - FORMULATION Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:10am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL LOW LEVEL-FIELD	SM 4500CL G	< 0.02 mg/l		05/01/09 11:10AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:43PM DH2
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:43PM DH2

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-3	DRINKING WATER - ROOM 48 - RACK RB21 Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:15am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL	SM 4500CL G	0.24 mg/l	0.02 mg/l	05/01/09 11:15AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:43PM DH2
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:43PM DH2

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-4	DRINKING WATER - ROOM 21 - RACK 109 Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:19am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL	SM 4500CL G	0.69 mg/l	0.02 mg/l	05/01/09 11:19AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:43PM DH2

Serial Number: 1100921

12 May 2011

1205 Industrial Blvd., P.O. Box 514, Southampton, PA 18966-0514 Phone: 215-355-3900 Fax: 215-355-7231 www.qclaboratories.com

EXACT COPY

MT 04 Jun 09

QC Laboratories

Analytical Report

ACCREDITED IN ACCORDANCE WITH nelac
05/05/09 02:23pm

Account No: WO5899, CHARLES RIVER LAB
Project No: WO5899, CHARLES RIVER LAB
P.O. No:
PWSID No:
Inv. No: 1088907

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-4	DRINKING WATER - ROOM 21 - RACK 109	05/01/09 11:19am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:43PM DH2

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-5	DRINKING WATER - FILL STATION Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:24am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL	SM 4500CL G	0.91 mg/l	0.02 mg/l	05/01/09 11:24AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:43PM DH2
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:43PM DH2

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-6	DRINKING WATER - ANALYTICAL Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:27am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL LOW LEVEL-FIELD	SM 4500CL G	< 0.02 mg/l		05/01/09 11:27AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:43PM DH2
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:43PM DH2

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2952712-7	DRINKING WATER - H-1 Received Temp: 38 F Iced (Y/N): Y	05/01/09 11:32am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL	SM 4500CL G	1.25 mg/l (1)	0.02 mg/l	05/01/09 11:32AM CU
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/01/09 04:43PM DH2
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/01/09 04:43PM DH2

L2952712-1:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L2952712-2:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

Serial Number: 1100921
Thomas J. Hines, President

(1) Internal chlorine checked and is within range, re test requested.
mw 12 may 2009

Approved
12 may 2009

EXACT COPY
MT 04 Jun 09

QC Laboratories

Analytical Report

ACCREDITED IN ACCORDANCE WITH nelac
05/05/09 02:23pm

Account No: W05899, CHARLES RIVER LAB P.O. No: Inv. No: 1088907
Project No: W05899, CHARLES RIVER LAB PWSID No:

L2952712-3:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L2952712-4:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L2952712-5:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L2952712-6:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L2952712-7:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

- A result of "ND" indicates the concentration of the analyte tested was either not detected or below the RLs.
- Definitions: ND=not detected; NEG=negative; POS=positive; COL=colonies; RLs=laboratory reporting limits; L/A=laboratory accident; TNTC=too numerous to count
- A result marked with "DRY" indicates that the result was calculated and reported on a dry weight basis.
- All analysis, except field tests are conducted in Southampton, PA unless otherwise identified.
- The test"pH lab"is analyzed upon receipt at the laboratory, the result will not be suitable for regulatory purposes.
- The reported results relate only to the samples.
- QC NELAP ID's:PA 09-00131,NJ PA166,FL E87954,NY 11223,CT PH-0768,DE PA-018,KY 90228,MD 206,EPA PA00018.Bioassay:PA 09-03574,NJ PA034,FL E87953,KS E10373,SC 89020001.
- QC STATE ID's:Wind Gap,NJ PA001,PA 48-01334;E RUTHERFORD NJ02015;Vineland NJ06005; Reading PA 06-03543.
- All samples are collected as "grab" samples unless otherwise identified.
- MCL= is the EPA recommended "maximum contaminant level" for a parameter. PLs=customer specific permit limits.
- The test results meet all requirements of NELAC unless otherwise specified.
- The report shall not be reproduced except in full without the written consent of the laboratory.

Regulatory authorities are assessing substantial fines for testing omissions. Please track your sample collections and results on a weekly, monthly, or quarterly basis to ensure compliance. QC's internet program 'LIVE ACCESS' will provide you with real-time access to collection dates and results. Please contact Customer Service for further information on acquiring LIVE ACCESS.

Page 3 of 3 Serial Number: 1100921

Approved
12 may 2009

EXACT COPY

MT 04 Jun 09

QC Laboratories

Analytical Report

ACCREDITED IN ACCORDANCE WITH nelac
05/15/09 12:07pm

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Regarding:

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1091338

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L2983332-1	DRINKING WATER - ROOM H1 Received Temp: 38 F Iced (Y/N): Y	05/13/09 01:25pm NA F	Theresa M. Boyle, QC Inc.

Parameter	Method	Result	RLs	Test Date, Time, Analyst
CHLORINE RESIDUAL	SM 4500CL G	0.4 mg/l	0.02 mg/l	05/13/09 01:25PM TMB
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	05/13/09 03:26PM AMD
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	05/13/09 03:26PM ARD

L2983332-1:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L2983332-1:
1. A water supply should not exceed the following limits: Coliform- <1 col/100ml or NEG; Iron-0.30 mg/l; Manganese-0.05 mg/l; Nitrate-10. mg/l; pH- 6.5 to 8.5 units.

2. Filtration system installed.

3. 09 15:26

- A result of "ND" indicates the concentration of the analyte tested was either not detected or below the RLs.
- Definitions: ND=not detected; NEG=negative; POS=positive; COL=colonies; RLs=laboratory reporting limits; L/A=laboratory accident; TNTC=too numerous to count
- A result marked with "DRY" indicates that the result was calculated and reported on a dry weight basis.
- All analysis, except field tests are conducted in Southampton, PA unless otherwise identified.
- The test"pH lab"is analyzed upon receipt at the laboratory, the result will not be suitable for regulatory purposes.
- The reported results relate only to the samples.
- QC NELAP ID's:PA 09-00131,NJ PA166,FL E87954,NY 11223,CT PH-0768,DE PA-018,KY 90228,MD 206,EPA PA00018.Bioassay:PA 09-03574,NJ PA034,FL E87953,KS E10373,SC 89020001.
- QC STATE ID's:Wind Gap,NJ PA001,PA 48-01334;E RUTHERFORD NJ02015;Vineland NJ06005; Reading PA 06-03543.
- All samples are collected as "grab" samples unless otherwise identified.

This report is a revision of report number 1107192
Serial Number: 1107409

Approved
19 may 2009

1205 Industrial Blvd., P.O. Box 514, Southampton, PA 18966-0514 Phone: 215-355-3900 Fax: 215-355-7231 www.qclaboratories.com

EXACT COPY
MPV 08/MAR/2010

QC Laboratories

Analytical Report

05/15/09 12:07pm

Account No: WO5899, CHARLES RIVER LAB
Project No: WO5899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1091338

- MCL= is the EPA recommended "maximum contaminant level" for a parameter. PLs=customer specific permit limits.
- The test results meet all requirements of NELAC unless otherwise specified.
- The report shall not be reproduced except in full without the written consent of the laboratory.

Regulatory authorities are assessing substantial fines for testing omissions. Please track your sample collections and results on a weekly, monthly, or quarterly basis to ensure compliance. QC's internet program 'LIVE ACCESS' will provide you with real-time access to collection dates and results. Please contact Customer Service for further information on acquiring LIVE ACCESS.

Approved
19 may 2009

This report is a revision of report number 1107192
Serial Number: 1107409

EXACT COPY
HPV 08 MAR 2010

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. 5576808 WW Group No. 1128104

Point #1 905 Analytical Lab Grab Water Sample
Semi-Annual

Collected:01/14/2009 09:55 by EA

Account Number: 02423

Submitted: 01/14/2009 15:30
Reported: 01/27/2009 at 20:43
Discard: 02/11/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

10905

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Units	Dilution Factor
00259	Mercury	7439-97-6	N.D.	0.000056	0.00020	mg/l	1
07035	Arsenic	7440-38-2	N.D.	0.0100	0.0200	mg/l	1
07036	Selenium	7782-49-2	N.D.	0.0107	0.0200	mg/l	1
07046	Barium	7440-39-3	N.D.	0.00060	0.0050	mg/l	1
07049	Cadmium	7440-43-9	N.D.	0.0020	0.0050	mg/l	1
07051	Chromium	7440-47-3	N.D.	0.0030	0.0150	mg/l	1
07055	Lead	7439-92-1	N.D.	0.0069	0.0150	mg/l	1
07066	Silver	7440-22-4	N.D.	0.0022	0.0050	mg/l	1
07072	Zinc	7440-66-6	N.D.	0.0081	0.0200	mg/l	1
00224	Chloride	16887-00-6	N.D.	1.0	2.0	mg/l	5
00226	Ortho-Phosphate as P	7723-14-0	N.D.	0.010	0.030	mg/l	1
00228	Sulfate	14808-79-8	N.D.	1.5	5.0	mg/l	5
00368	Nitrate Nitrogen	14797-55-8	N.D.	0.25	0.50	mg/l	5
01504	Fluoride	16984-48-8	N.D.	0.40	0.50	mg/l	5
01505	Bromide	24959-67-9	N.D.	2.0	2.5	mg/l	5
01506	Nitrite Nitrogen	14797-65-0	N.D.	0.40	0.50	mg/l	5
01856	Herbicides in Water						
01857	2,4-D	94-75-7	N.D.	0.15	0.48	ug/l	1
01858	2,4,5-TP	93-72-1	N.D.	0.0096	0.048	ug/l	1
05286	2,4,5-T	93-76-5	N.D.	0.014	0.048	ug/l	1
05287	Dalapon	75-99-0	N.D.	0.24	1.2	ug/l	1
05288	Dinoseb	88-85-7	N.D.	0.096	0.48	ug/l	1
05289	Dicamba	1918-00-9	N.D.	0.077	0.29	ug/l	1
05290	MCPP	93-65-2	N.D.	48	190	ug/l	1
05291	MCPA	94-74-6	N.D.	290	960	ug/l	1
05292	2,4-DP (Dichlorprop)	120-36-5	N.D.	0.15	0.48	ug/l	1
05293	2,4-DB	94-82-6	N.D.	0.29	0.96	ug/l	1
08103	Pentachlorophenol	87-86-5	N.D.	0.026	0.048	ug/l	1
00178	Pesticides/PCB's in Water						
00453	Gamma BHC - Lindane	58-89-9	N.D.	0.0045	0.0097	ug/l	1
00454	Heptachlor	76-44-8	N.D.	0.0039	0.0097	ug/l	1
00455	Aldrin	309-00-2	N.D.	0.0041	0.019	ug/l	1
00469	Dieldrin	60-57-1	N.D.	0.0039	0.019	ug/l	1
00477	Endrin	72-20-8	N.D.	0.0039	0.019	ug/l	1
00478	p,p-DDT	50-29-3	N.D.	0.0058	0.019	ug/l	1

EXACT COPY
MT 04 JUN 09

Approved
29 JAN 2009

*=This limit was used in the evaluation of the final result

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax:717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. 5576808 WW Group No. 1128104

Point #1 905 Analytical Lab Grab Water Sample
Semi-Annual

Collected:01/14/2009 09:55 by EA

Account Number: 02423

Submitted: 01/14/2009 15:30
Reported: 01/27/2009 at 20:43
Discard: 02/11/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

10905

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Units	Dilution Factor
00638	Endrin Aldehyde	7421-93-4	N.D.	0.019	0.097	ug/l	1
01902	Alpha BHC	319-84-6	N.D.	0.0026	0.0097	ug/l	1
01903	Beta BHC	319-85-7	N.D.	0.018	0.058	ug/l	1
01904	Delta BHC	319-86-8	N.D.	0.0041	0.0097	ug/l	1
01905	Heptachlor Epoxide	1024-57-3	N.D.	0.0029	0.0097	ug/l	1
01906	p,p-DDE	72-55-9	N.D.	0.0048	0.019	ug/l	1
01907	p,p-DDD	72-54-8	N.D.	0.0039	0.019	ug/l	1
01908	Chlordane	57-74-9	N.D.	0.068	0.48	ug/l	1
01909	Toxaphene	8001-35-2	N.D.	0.29	0.97	ug/l	1
01910	Endosulfan I	959-98-8	N.D.	0.0029	0.0097	ug/l	1
01911	Endosulfan II	33213-65-9	N.D.	0.0039	0.019	ug/l	1
01912	Endosulfan Sulfate	1031-07-8	N.D.	0.0039	0.019	ug/l	1
01913	PCB-1016	12674-11-2	N.D.	0.097	0.48	ug/l	1
01914	PCB-1221	11104-28-2	N.D.	0.15	0.48	ug/l	1
01915	PCB-1232	11141-16-5	N.D.	0.097	0.48	ug/l	1
01916	PCB-1242	53469-21-9	N.D.	0.097	0.48	ug/l	1
01917	PCB-1248	12672-29-6	N.D.	0.097	0.48	ug/l	1
01918	PCB-1254	11097-69-1	N.D.	0.097	0.48	ug/l	1
01919	PCB-1260	11096-82-5	N.D.	0.097	0.48	ug/l	1

EXACT COPY

04 Jun 09

PA DEP Lab Certification ID 36-00037, Expiration Date: 1/31/09

All QC is compliant unless otherwise noted. Please refer to the Quality Control Summary for overall QC performance data and associated samples.

Approved
29 JAN 2009

Laboratory Chronicle

CAT No.	Analysis Name	Method	Trial#	Analysis Date and Time	Analyst	Dilution Factor
00259	Mercury	EPA 245.1 rev 3	1	01/16/2009 07:13	Damary Valentin	1
07035	Arsenic	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
07036	Selenium	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
07046	Barium	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
07049	Cadmium	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
07051	Chromium	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
07055	Lead	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
07066	Silver	EPA 200.7 rev 4.4	1	01/23/2009 20:44	Thomas F McLamb Sr	1

*=This limit was used in the evaluation of the final result

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. 5576808 WW Group No. 1128104

Point #1 905 Analytical Lab Grab Water Sample
Semi-Annual

Collected:01/14/2009 09:55 by EA

Account Number: 02423

Submitted: 01/14/2009 15:30
Reported: 01/27/2009 at 20:43
Discard: 02/11/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

10905

07072	Zinc	EPA 200.7 rev 4.4	1	01/22/2009 23:11	Thomas F McLamb Sr	1
00224	Chloride	EPA 300.0	1	01/15/2009 09:33	Ashley M Heckman	5
00226	Ortho-Phosphate as P	EPA 365.3	1	01/14/2009 21:35	Daniel S Smith	1
00228	Sulfate	EPA 300.0	1	01/15/2009 09:33	Ashley M Heckman	5
00368	Nitrate Nitrogen	EPA 300.0	1	01/15/2009 09:33	Ashley M Heckman	5
01504	Fluoride	EPA 300.0	1	01/17/2009 02:16	Ashley M Heckman	5
01505	Bromide	EPA 300.0	1	01/15/2009 09:33	Ashley M Heckman	5
01506	Nitrite Nitrogen	EPA 300.0	1	01/15/2009 09:33	Ashley M Heckman	5
01856	Herbicides in Water	SW-846 8151A	1	01/16/2009 19:07	Tricia M Gusbar	1
00178	Pesticides/PCB's in Water	EPA 608	1	01/17/2009 00:44	Lindsey K Lafferty	1
00816	Water Sample Herbicide Extract	SW-846 8151A	1	01/15/2009 22:00	Olivia I Santiago	1
05714	PW/WW Hg Digest	EPA 245.1 rev 3	1	01/15/2009 20:00	Nelli S Markaryan	1
05716	EPA 600 ICP Digest (tot rec)	EPA 200.7 rev 4.4	1	01/22/2009 09:03	Denise K Conners	1
10241	Method 608 Water Extraction	EPA 608	1	01/16/2009 03:00	Sherry L Morrow	1

Approved
29 JAN2009

EXACT COPY
MT 04 Jun 09

*=This limit was used in the evaluation of the final result

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. 5576809 WW Group No. 1128104

Point #2 905 Formulation Lab Grab Water Sample
Semi-Annual

Collected:01/14/2009 10:05 by EA

Account Number: 02423

Submitted: 01/14/2009 15:30
Reported: 01/27/2009 at 20:43
Discard: 02/11/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

20905

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Units	Dilution Factor
00259	Mercury	7439-97-6	N.D.	0.000056	0.00020	mg/l	1
07035	Arsenic	7440-38-2	N.D.	0.0100	0.0200	mg/l	1
07036	Selenium	7782-49-2	N.D.	0.0107	0.0200	mg/l	1
07046	Barium	7440-39-3	N.D.	0.00060	0.0050	mg/l	1
07049	Cadmium	7440-43-9	N.D.	0.0020	0.0050	mg/l	1
07051	Chromium	7440-47-3	N.D.	0.0030	0.0150	mg/l	1
07055	Lead	7439-92-1	N.D.	0.0069	0.0150	mg/l	1
07066	Silver	7440-22-4	N.D.	0.0022	0.0050	mg/l	1
07072	Zinc	7440-66-6	N.D.	0.0081	0.0200	mg/l	1
00224	Chloride	16887-00-6	1.2 J	1.0	2.0	mg/l	5
00226	Ortho-Phosphate as P	7723-14-0	N.D.	0.010	0.030	mg/l	1
00228	Sulfate	14808-79-8	N.D.	1.5	5.0	mg/l	5
00368	Nitrate Nitrogen	14797-55-8	N.D.	0.25	0.50	mg/l	5
01504	Fluoride	16984-48-8	N.D.	0.40	0.50	mg/l	5
01505	Bromide	24959-67-9	N.D.	2.0	2.5	mg/l	5
01506	Nitrite Nitrogen	14797-65-0	N.D.	0.40	0.50	mg/l	5
01856	Herbicides in Water						
01857	2,4-D	94-75-7	N.D.	0.16	0.51	ug/l	1
01858	2,4,5-TP	93-72-1	N.D.	0.010	0.051	ug/l	1
05286	2,4,5-T	93-76-5	N.D.	0.015	0.051	ug/l	1
05287	Dalapon	75-99-0	N.D.	0.25	1.3	ug/l	1
05288	Dinoseb	88-85-7	N.D.	0.10	0.51	ug/l	1
05289	Dicamba	1918-00-9	N.D.	0.081	0.30	ug/l	1
05290	MCPP	93-65-2	N.D.	51	200	ug/l	1
05291	MCPA	94-74-6	N.D.	300	1,000	ug/l	1
05292	2,4-DP (Dichlorprop)	120-36-5	N.D.	0.16	0.51	ug/l	1
05293	2,4-DB	94-82-6	N.D.	0.30	1.0	ug/l	1
08103	Pentachlorophenol	87-86-5	N.D.	0.027	0.051	ug/l	1
00178	Pesticides/PCB's in Water						
00453	Gamma BHC - Lindane	58-89-9	N.D.	0.0044	0.0096	ug/l	1
00454	Heptachlor	76-44-8	N.D.	0.0038	0.0096	ug/l	1
00455	Aldrin	309-00-2	N.D.	0.0040	0.019	ug/l	1
00469	Dieldri	60-57-1	N.D.	0.0038	0.019	ug/l	1
00477	Endrin	72-20-8	N.D.	0.0038	0.019	ug/l	1
00478	p,p-DDT	50-29-3	N.D.	0.0058	0.019	ug/l	1

*=This limit was used in the evaluation of the final result

EXACT COPY
HPV 08 | MAR | 2010

Approved
29 May 2009

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 •717-656-2300 Fax:717-656-2681• www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. 5576809 WW Group No. 1128104

Point #2 905 Formulation Lab Grab Water Sample
Semi-Annual

Collected:01/14/2009 10:05 by EA

Account Number: 02423

Submitted: 01/14/2009 15:30
Reported: 01/27/2009 at 20:43
Discard: 02/11/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

20905

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Units	Dilution Factor
00638	Endrin Aldehyde	7421-93-4	N.D.	0.019	0.096	ug/l	1
01902	Alpha BHC	319-84-6	N.D.	0.0026	0.0096	ug/l	1
01903	Beta BHC	319-85-7	N.D.	0.018	0.058	ug/l	1
01904	Delta BHC	319-86-8	N.D.	0.0040	0.0096	ug/l	1
01905	Heptachlor Epoxide	1024-57-3	N.D.	0.0029	0.0096	ug/l	1
01906	p,p-DDE	72-55-9	N.D.	0.0048	0.019	ug/l	1
01907	p,p-DDD	72-54-8	N.D.	0.0038	0.019	ug/l	1
01908	Chlordane	57-74-9	N.D.	0.067	0.48	ug/l	1
01909	Toxaphene	8001-35-2	N.D.	0.29	0.96	ug/l	1
01910	Endosulfan I	959-98-8	N.D.	0.0029	0.0096	ug/l	1
01911	Endosulfan II	33213-65-9	N.D.	0.0038	0.019	ug/l	1
01912	Endosulfan Sulfate	1031-07-8	N.D.	0.0038	0.019	ug/l	1
01913	PCB-1016	12674-11-2	N.D.	0.096	0.48	ug/l	1
01914	PCB-1221	11104-28-2	N.D.	0.15	0.48	ug/l	1
01915	PCB-1232	11141-16-5	N.D.	0.096	0.48	ug/l	1
01916	PCB-1242	53469-21-9	N.D.	0.096	0.48	ug/l	1
01917	PCB-1248	12672-29-6	N.D.	0.096	0.48	ug/l	1
01918	PCB-1254	11097-69-1	N.D.	0.096	0.48	ug/l	1
01919	PCB-1260	11096-82-5	N.D.	0.096	0.48	ug/l	1

PA DEP Lab Certification ID 36-00037, Expiration Date: 1/31/09

All QC is compliant unless otherwise noted. Please refer to the Quality Control Summary for overall QC performance data and associated samples.

Laboratory Chronicle

CAT No.	Analysis Name	Method	Trial#	Analysis Date and Time	Analyst	Dilution Factor
00259	Mercury	EPA 245.1 rev 3	1	01/16/2009 07:14	Damary Valentin	1
07035	Arsenic	EPA 200.7 rev 4.4	1	01/23/2009 23:36	Thomas F McLamb Sr	1
07036	Selenium	EPA 200.7 rev 4.4	1	01/22/2009 22:21	John P Hook	1
07046	Barium	EPA 200.7 rev 4.4	1	01/22/2009 22:21	John P Hook	1
07049	Cadmium	EPA 200.7 rev 4.4	1	01/22/2009 22:21	John P Hook	1
07051	Chromium	EPA 200.7 rev 4.4	1	01/22/2009 22:21	John P Hook	1
07055	Lead	EPA 200.7 rev 4.4	1	01/22/2009 22:21	John P Hook	1
07066	Silver	EPA 200.7 rev 4.4	1	01/22/2009 22:21	John P Hook	1

*=This limit was used in the evaluation of the final result

EXACT COPY
HP V 08 | MAR | 2010

Approved
28 may 2009

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 •717-656-2300 Fax:717-656-2681• www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. 5576809 WW Group No. 1128104

Point #2 905 Formulation Lab Grab Water Sample
Semi-Annual

Collected:01/14/2009 10:05 by EA

Account Number: 02423

Submitted: 01/14/2009 15:30
Reported: 01/27/2009 at 20:43
Discard: 02/11/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

20905

07072	Zinc	EPA 200.7 rev 4.4	1	01/23/2009 23:36	Thomas F McLamb Sr	1
00224	Chloride	EPA 300.0	1	01/15/2009 09:49	Ashley M Heckman	5
00226	Ortho-Phosphate as P	EPA 365.3	1	01/14/2009 21:35	Daniel S Smith	1
00228	Sulfate	EPA 300.0	1	01/15/2009 09:49	Ashley M Heckman	5
00368	Nitrate Nitrogen	EPA 300.0	1	01/15/2009 09:49	Ashley M Heckman	5
01504	Fluoride	EPA 300.0	1	01/17/2009 02:32	Ashley M Heckman	5
01505	Bromide	EPA 300.0	1	01/15/2009 09:49	Ashley M Heckman	5
01506	Nitrite Nitrogen	EPA 300.0	1	01/15/2009 09:49	Ashley M Heckman	5
01856	Herbicides in Water	SW-846 8151A	1	01/16/2009 19:34	Tricia M Gusbar	1
00178	Pesticides/PCB's in Water	EPA 608	1	01/19/2009 11:29	Lindsey K Lafferty	1
00816	Water Sample Herbicide Extract	SW-846 8151A	1	01/15/2009 22:00	Olivia I Santiago	1
05714	PW/WW Hg Digest	EPA 245.1 rev 3	1	01/15/2009 20:00	Nelli S Markaryan	1
05716	EPA 600 ICP Digest (tot rec)	EPA 200.7 rev 4.4	1	01/22/2009 09:10	Denise K Conners	1
10241	Method 608 Water Extraction	EPA 608	1	01/16/2009 03:00	Sherry L Morrow	1

Approved
28 May 2009

*=This limit was used in the evaluation of the final result

EXACT COPY
MPV 08 MAR 2010

Lancaster Laboratories
Explanation of Symbols and Abbreviations

The following defines common symbols and abbreviations used in reporting technical data:

N.D.	none detected	**BMQL**	Below Minimum Quantitation Level
TNTC	Too Numerous To Count	**MPN**	Most Probable Number
IU	International Units	**CP Units**	cobalt-chloroplatinate units
umhos/cm	micromhos/cm	**NTU**	nephelometric turbidity units
C	degrees Celsius	**F**	degrees Fahrenheit
Cal	(diet) calories	**lb.**	pound(s)
meq	milliequivalents	**kg**	kilogram(s)
g	gram(s)	**mg**	milligram(s)
ug	microgram(s)	**l**	liter(s)
ml	milliliter(s)	**ul**	microliter(s)
m3	cubic meter(s)	**fib >5 um/ml**	fibers greater than 5 microns in length per ml

< less than – The number following the sign is the limit of quantitation, the smallest amount of analyte which can be reliably determined using this specific test.

> greater than

ppm parts per million – One ppm is equivalent to one milligram per kilogram (mg/kg), or one gram per million grams. For aqueous liquids, ppm is usually taken to be equivalent to milligrams per liter (mg/l), because one liter of water has a weight very close to a kilogram. For gases or vapors, one ppm is equivalent to one microliter of gas per liter of gas.

ppb parts per billion

Dry weight basis Results printed under this heading have been adjusted for moisture content. This increases the analyte weight concentration to approximate the value present in a similar sample without moisture.

U.S. EPA data qualifiers:

Organic Qualifiers

A	TIC is a possible aldol-condensation product
B	Analyte was also detected in the blank
C	Pesticide result confirmed by GC/MS
D	Compound quatitated on a diluted sample
E	Concentration exceeds the calibration range of the instrument
J	Estimated value
N	Presumptive evidence of a compound (TICs only)
P	Concentration difference between primary and confirmation columns >25%
U	Compound was not detected
X,Y,Z	Defined in case narrative

Inorganic Qualifiers

B	Value is <CRDL, but ≥IDL
E	Estimated due to interference
M	Duplicate injection precision not met
N	Spike amount not within control limits
S	Method of standard additions (MSA) used for calculation
U	Compound was not detected
W	Post digestion spike out of control limits
*****	Duplicate analysis not within control limits
+	Correlation coefficient for MSA <0.995

EXACT COPY

MT 04 Jun 09

Analytical test results for methods listed on the laboratories' accreditation scope meet all requirements of NELAC unless otherwise noted under the individual analysis.

Tests results relate only to the sample tested. Clients should be aware that a critical step in a chemical or microbiological analysis is the collection of the sample. Unless the sample analyzed is truly representative of the bulk of material involved, the test results will be meaningless. If you have questions regarding the proper techniques of collecting samples, please contact us. We cannot be held responsible for sample integrity, however, unless sampling has been performed by a member of our staff. This report shall not be reproduced except in full, without the written approval of the laboratory.

WARRANTY AND LIMITS OF LIABILITY – In accepting analytical work, we warrant the accuracy of test results for the sample as submitted. THE FOREGOING EXPRESS WARRANTY IS EXCLUSIVE AND IS GIVEN IN LIEU OF ALL OTHER WARRANTIES, EXPRESSED OR IMPLIED. WE DISCLAIM ANY OTHER WARRANTIES, EXPRESSED OR IMPLIED, INCLUDING A WARRANTY OF FITNESS FOR PARTICULAR PURPOSE AND WARRANTY OF MERCHANTABILITY. IN NO EVENT SHALL LANCASTER LABORATORIES BE LIABLE FOR INDIRECT, SPECIAL, CONSEQUENTIAL, OR INCIDENTAL DAMAGES INCLUDING, BUT NOT LIMITED TO, DAMAGES FOR LOSS OF PROFIT OR GOODWILL REGARDLESS OF (A) THE NEGLIGENCE (EITHER SOLE OR CONCURRENT) OF LANCASTER LABORATORIES AND (B) WHETHER LANCASTER LABORATORIES HAS BEEN INFORMED OF THE POSSIBILITY OF SUCH DAMAGES. We accept no legal responsibility for the purposes for which the client uses the test results. No purchase order or other order for work shall be accepted by Lancaster Laboratories which includes any conditions that vary from the Standard Terms and Conditions of Lancaster Laboratories and we hereby object to any conflicting terms contained in any acceptance or order submitted by client.

QC Laboratories

Analytical Report

11/11/09 10:01am

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Regarding:

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1146923

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-1	DRINKING WATER - IN VITRO Received Temp: 38 F Iced (Y/N): Y	11/06/09 11:32am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL LOW LEVEL-FIELD	SM 4500CL G	< 0.02 mg/l		11/06/09 11:32AM CU

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-2	DRINKING WATER - ANALYTICAL Received Temp: 38 F Iced (Y/N): Y	11/06/09 11:47am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL LOW LEVEL-FIELD	SM 4500CL G	< 0.02 mg/l		11/06/09 11:47AM CU

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-3	DRINKING WATER - FILLING STATION Received Temp: 38 F Iced (Y/N): Y	11/06/09 11:53am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF

Serial Number: 1245466

Approved
07 Dec 09

1205 Industrial Blvd., P.O. Box 514, Southampton, PA 18966-0514 Phone: 215-355-3900 Fax: 215-355-7231 www.qclaboratories.com

EXACT COPY
HPJ 22 APR 2010

QC Laboratories

Analytical Report

11/11/09 10:01am

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB
P.O. No:
PWSID No:
Inv. No: 1146923

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-3	DRINKING WATER - FILLING STATION	11/06/09 11:53am NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL	SM 4500CL G	1.19 mg/l	0.02 mg/l	11/06/09 11:53AM CU

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-4	DRINKING WATER - ROOM 1 RACK 123 Received Temp: 38 F Iced (Y/N): Y	11/06/09 12:09pm NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL	SM 4500CL G	0.97 mg/l	0.02 mg/l	11/06/09 12:09PM CU

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-5	DRINKING WATER - ROOM 51 RACK 042 Received Temp: 38 F Iced (Y/N): Y	11/06/09 12:16pm NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL	SM 4500CL G	5.9 mg/l	0.02 mg/l	11/06/09 12:16PM CU

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-6	DRINKING WATER - FORMULATION Received Temp: 38 F Iced (Y/N): Y	11/06/09 12:24pm NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL LOW LEVEL-FIELD	SM 4500CL G	< 0.02 mg/l		11/06/09 12:24PM CU

Approved
07 Dec 09

Serial Number: 1245466

Thomas J. Hines, President

EXACT COPY
MPV 22 APR 2010

QC Laboratories

Analytical Report

11/11/09 10:01am

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1146923

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3151048-7	DRINKING WATER - H-2 Received Temp: 38 F Iced (Y/N): Y	11/06/09 12:05pm NA F	Customer Sampled

Parameter	Method	Result	RLs	Test Date, Time, Analyst
ENVIRONMENTAL MICROBIOLOGY				
COLIFORM-MF	SM 9222B	<1 col/100ml	1. col/100ml	11/07/09 01:03PM TF
STANDARD PLATE COUNT	SM 9215B	<1 col/ml	1. col/ml	11/07/09 01:03PM TF
FIELD SERVICES				
CHLORINE RESIDUAL	SM 4500CL G	0.6 mg/l	0.02 mg/l	11/06/09 12:05PM CU

L3151048-1:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L3151048-2:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L3151048-3:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L3151048-4:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L3151048-5:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

Page 3 of 4

Serial Number: 1245466

Approved
07DEC09

Thomas J. Hines, President

EXACT COPY
MPV 22 APR 2010

QC Laboratories

Analytical Report

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1146923

L3151048-6:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

L3151048-7:
1. A water supply is considered bacteriologically "SAFE" if no Coliform bacteria are detected. To be considered "SAFE" your report should indicate "<1 col/100ml" or "NEG" for the Coliform Test. If your report indicates a positive result "POS" or a value of one (1) or greater then your supply is "UNSAFE FOR DRINKING" contact your local Health Dept. or QC for advice.

- A result of "ND" indicates the concentration of the analyte tested was either not detected or below the RLs.
- Definitions: ND=not detected; NEG=negative; POS=positive; COL=colonies; RLs=laboratory reporting limits; L/A=laboratory accident; TNTC=too numerous to count
- A result marked with "DRY" indicates that the result was calculated and reported on a dry weight basis.
- All analysis, except field tests are conducted in Southampton, PA unless otherwise identified.
- The test"pH lab"is analyzed upon receipt at the laboratory, the result will not be suitable for regulatory purposes.
- The reported results relate only to the samples.
- QC NELAP ID's:PA 09-00131,NJ PA166,FL E87954,NY 11223,CT PH-0768,DE PA-018,KY 90228,MD 206,EPA PA00018.Bioassay:PA 09-03574,NJ PA034,FL E87953,KS E10373,SC 89020001.
- QC STATE ID's:Wind Gap,NJ PA001,PA 48-01334;E RUTHERFORD NJ02015;Vineland NJ06005; Reading PA 06-03543.
- All samples are collected as "grab" samples unless otherwise identified.
- MCL= is the EPA recommended "maximum contaminant level" for a parameter. PLs=customer specific permit limits.
- The test results meet all requirements of NELAC unless otherwise specified.
- The report shall not be reproduced except in full without the written consent of the laboratory.

Regulatory authorities are assessing substantial fines for testing omissions. Please track your sample collections and results on a weekly, monthly, or quarterly basis to ensure compliance. QC's internet program 'LIVE ACCESS' will provide you with real-time access to collection dates and results. Please contact Customer Service for further information on acquiring LIVE ACCESS.

Approved
07Dec09

Page 4 of 4

Serial Number: 1245466

Thomas J. Hines, President

EXACT COPY
HPV 22 APR 2010

QC Laboratories

Analytical Report

11/12/09 10:22am

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Regarding:

MATTHEW VANEMAN
CHARLES RIVER LAB
905 SHEEHY DRIVE
HORSHAM, PA 19044

Account No: W05899, CHARLES RIVER LAB
Project No: W05899, CHARLES RIVER LAB

P.O. No:
PWSID No:

Inv. No: 1147178

Sample Number	Sample Description	Samp. Date/Time/Temp	Sampled by
L3172200-1	ROOM 51 RACK 042	11/09/09 09:50am NA F	Joan Cummings Nulty, QC Laboratories

Parameter	Method	Result	RLs	Test Date, Time, Analyst
FIELD SERVICES				
CHLORINE RESIDUAL	SM 4500CL G	0.69 mg/l	0.02 mg/l	11/09/09 09:50AM JCN

- A result of "ND" indicates the concentration of the analyte tested was either not detected or below the RLs.
- Definitions: ND=not detected; NEG=negative; POS=positive; COL=colonies; RLs=laboratory reporting limits; L/A=laboratory accident; TNTC=too numerous to count
- A result marked with "DRY" indicates that the result was calculated and reported on a dry weight basis.
- All analysis, except field tests are conducted in Southampton, PA unless otherwise identified.
- The test"pH lab"is analyzed upon receipt at the laboratory, the result will not be suitable for regulatory purposes.
- The reported results relate only to the samples.
- QC NELAP ID's:PA 09-00131,NJ PA166,FL E87954,NY 11223,CT PH-0768,DE PA-018,KY 90228,MD 206,EPA PA00018.Bioassay:PA 09-03574,NJ PA034,FL E87953,KS E10373,SC 89020001.
- QC STATE ID's:Wind Gap,NJ PA001,PA 48-01334;E RUTHERFORD NJ02015;Vineland NJ06005; Reading PA 06-03543.
- All samples are collected as "grab" samples unless otherwise identified.
- MCL= is the EPA recommended "maximum contaminant level" for a parameter. PLs=customer specific permit limits.
- The test results meet all requirements of NELAC unless otherwise specified.
- The report shall not be reproduced except in full without the written consent of the laboratory.

Regulatory authorities are assessing substantial fines for testing omissions. Please track your sample collections and results on a weekly, monthly, or quarterly basis to ensure compliance. QC's internet program 'LIVE ACCESS' will provide you with real-time access to collection dates and results. Please contact Customer Service for further information on acquiring LIVE ACCESS.

Approved
07 Dec 09

Page 1 of 1

Serial Number: 1246275

Thomas J. Hines, President

1205 Industrial Blvd., P.O. Box 514, Southampton, PA 18966-0514 Phone: 215-355-3900 Fax: 215-355-7231 www.qclaboratories.com

EXACT copy
MPV 22/APR/2010

Memo

Date: 10 December 2009

To: Water Analysis File 10Dec2009

From: Matthew J. Vaneman, Director of Operation

RE: Over Chlorination of R.O. System #2

On November 6th 2009, QC Laboratories performed testing of the R.O. System in 905 F. The results of the analysis indicated a high level of chlorine. All animal racks were unhooked from the system. The system was drained and refilled with water from R.O. System #1. The system and racks were flushed with the water from R.O. System #1 and were maintained by adding water from R.O. System #1 to the R.O. System #2 storage tank until the system repair was made. All water in each rack was tested for chlorine content at the time of discovery (results attached). A non-routine maintenance form was completed for the R.O. System. The system was evaluated and repaired. Further details can be found in the maintenance records.

EXACT COPY
HPV 22 APR 2010

Lancaster Laboratories — Analysis Report

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Lancaster Laboratories Sample No. WW 5712406 Group No. 1151471
PA

Sample #2 905 Formulation Lab Grab Water Sample
Semi-Annual

Collected: 06/30/2009 09:56 by JF

Account Number: 02423

Submitted: 06/30/2009 16:30
Reported: 07/13/2009 at 14:33
Discard: 07/28/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

20905

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Dilution Factor
SW-846 8151A	**Herbicides**		ug/l	ug/l	ug/l	
01856	2,4-D	94-75-7	N.D.	0.16	0.50	1
01856	Dalapon	75-99-0	N.D.	0.25	1.3	1
01856	2,4-DB	94-82-6	N.D.	0.30	1.0	1
01856	Dicamba	1918-00-9	N.D.	0.080	0.30	1
01856	Dinoseb	88-85-7	N.D.	0.10	0.50	1
01856	2,4-DP (Dichlorprop)	120-36-5	N.D.	0.16	0.50	1
01856	MCPA	94-74-6	N.D.	300	1,000	1
1856	MCPP	93-65-2	380 (1)	50	200	1
356	Pentachlorophenol	87-86-5	N.D.	0.027	0.050	1
1856	2,4,5-T	93-76-5	N.D.	0.015	0.050	1
01856	2,4,5-TP	93-72-1	N.D.	0.010	0.050	1
EPA 608	**Pesticides/PCBs**		ug/l	ug/l	ug/l	
00178	Aldrin	309-00-2	N.D.	0.0040	0.019	1
00178	Alpha BHC	319-84-6	N.D.	0.0026	0.0096	1
00178	Beta BHC	319-85-7	N.D.	0.018	0.057	1
00178	Gamma BHC - Lindane	58-89-9	N.D.	0.0044	0.0096	1
00178	Chlordane	57-74-9	N.D.	0.067	0.48	1
00178	p,p-DDD	72-54-8	N.D.	0.0038	0.019	1
00178	p,p-DDE	72-55-9	N.D.	0.0048	0.019	1
00178	p,p-DDT	50-29-3	N.D.	0.0057	0.019	1
00178	Delta BHC	319-86-8	N.D.	0.0040	0.0096	1
00178	Dieldrin	60-57-1	N.D.	0.0038	0.019	1
00178	Endosulfan I	959-98-8	N.D.	0.0029	0.0096	1
00178	Endosulfan II	33213-65-9	N.D.	0.0038	0.019	1
00178	Endosulfan Sulfate	1031-07-8	N.D.	0.0038	0.019	1
00178	Endrin	72-20-8	N.D.	0.0038	0.019	1
00178	Endrin Aldehyde	7421-93-4	N.D.	0.019	0.096	1
00178	Heptachlor	76-44-8	N.D.	0.0038	0.0096	1
00178	Heptachlor Epoxide	1024-57-3	N.D.	0.0029	0.0096	1
00178	PCB-1016	12674-11-2	N.D.	0.096	0.48	1
00178	PCB-1221	11104-28-2	N.D.	0.15	0.48	1
00178	PCB-1232	11141-16-5	N.D.	0.096	0.48	1
00178	PCB-1242	53469-21-9	N.D.	0.096	0.48	1
00178	PCB-1248	12672-29-6	N.D.	0.096	0.48	1
00178	PCB-1254	11097-69-1	N.D.	0.096	0.48	1
00178	PCB-1260	11096-82-5	N.D.	0.096	0.48	1
00178	Toxaphene	8001-35-2	N.D.	0.29	0.96	1
EPA 200.7 rev 4.4	**Metals**		mg/l	mg/l	mg/l	
07035	Arsenic	7440-38-2	N.D.	0.0072	0.0200	1
07046	Barium	7440-39-3	N.D.	0.00060	0.0050	1
07049	Cadmium	7440-43-9	N.D.	0.0020	0.0050	1
7051	Chromium	7440-47-3	N.D.	0.0034	0.0150	1
055	Lead	7439-92-1	N.D.	0.0069	0.0150	1
036	Selenium	7782-49-2	N.D.	0.0089	0.0200	1
07066	Silver	7440-22-4	N.D.	0.0023	0.0050	1

*This limit was used in the evaluation of the final result

(1) No maximum limit has been established by EPA. Amount here is ~~minimal~~ minimal appears to be of no consequence. AH 6 Aug 09 Spelling clarity AH

Approved [signature] 21 Jul 2009

EXACT COPY
[initials] 22 APR 2010

Lancaster Laboratories
2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. WW 5712406 **Group No. 1151471**
PA

Sample #2 905 Formulation Lab Grab Water Sample
Semi-Annual

Collected: 06/30/2009 09:56 by JF

Account Number: 02423

Submitted: 06/30/2009 16:30
Reported: 07/13/2009 at 14:33
Discard: 07/28/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

20905

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Dilution Factor
EPA 200.7 rev 4.4	**Metals**		mg/l	mg/l	mg/l	
07072	Zinc	7440-66-6	N.D.	0.0081	0.0200	1
EPA 245.1 rev 3	**Metals**		mg/l	mg/l	mg/l	
00259	Mercury	7439-97-6	N.D.	0.000056	0.00020	1
EPA 300.0	**Wet Chemistry**		mg/l	mg/l	mg/l	
01505	Bromide	24959-67-9	N.D.	2.0	2.5	5
224	Chloride	16887-00-6	N.D.	1.0	2.0	5
504	Fluoride	16984-48-8	N.D.	0.40	0.50	5
00368	Nitrate Nitrogen	14797-55-8	N.D.	0.25	0.50	5
01506	Nitrite Nitrogen	14797-65-0	N.D.	0.40	0.50	5
00228	Sulfate	14808-79-8	N.D.	1.5	5.0	5
EPA 365.3	**Wet Chemistry**		mg/l	mg/l	mg/l	
00226	Ortho-Phosphate as P	7723-14-0	N.D.	0.010	0.030	1

General Sample Comments

PA DEP Lab Certification ID 36-00037, Expiration Date: 1/31/10

All QC is compliant unless otherwise noted. Please refer to the Quality Control Summary for overall QC performance data and associated samples.

Laboratory Sample Analysis Record

CAT No.	Analysis Name	Method	Trial#	Batch#	Analysis Date and Time	Analyst	Dilution Factor
01856	Herbicides in Water	SW-846 8151A	1	091820017A	07/06/2009 16:23	John W Perkins	1
00178	Pesticides/PCB's in Water	EPA 608	1	091820005A	07/09/2009 02:08	Mark E McNulty	1
10241	Method 608 Water Extraction	EPA 608	1	091820005A	07/01/2009 17:00	JoElla L Rice	1
00816	Water Sample Herbicide Extract	SW-846 8151A	1	091820017A	07/02/2009 01:00	Tracy L Schickel	1
07035	Arsenic	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
07046	Barium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
07049	Cadmium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
07051	Chromium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
07055	Lead	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
07036	Selenium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 18:55	John P Hook	1
07066	Silver	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
07072	Zinc	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:54	Tara L Snyder	1
259	Mercury	EPA 245.1 rev 3	1	091875714003	07/09/2009 17:26	Parker D Lindstrom	1
716	EPA 600 ICP Digest (tot rec)	EPA 200.7 rev 4.4	1	091875716006	07/08/2009 14:26	James L Mertz	1

*This limit was used in the evaluation of the final result

Approved
MAD
21-Jul 2009

EXACT COPY
HPV 22 APR 2016

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. WW 5712406

Group No. 1151471
PA

Sample #2 905 Formulation Lab Grab Water Sample
Semi-Annual

Collected: 06/30/2009 09:56 by JF

Account Number: 02423

Submitted: 06/30/2009 16:30
Reported: 07/13/2009 at 14:33
Discard: 07/28/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

20905

Laboratory Sample Analysis Record

CAT No.	Analysis Name	Method	Trial#	Batch#	Analysis Date and Time	Analyst	Dilution Factor
05714	PW/WW Hg Digest	EPA 245.1 rev 3	1	091875714003	07/08/2009 15:30	James L Mertz	1
01505	Bromide	EPA 300.0	1	09182196601A	07/01/2009 12:43	Ashley M Adams	5
00224	Chloride	EPA 300.0	1	09182196601A	07/01/2009 12:43	Ashley M Adams	5
01504	Fluoride	EPA 300.0	1	09182196601A	07/01/2009 12:43	Ashley M Adams	5
00368	Nitrate Nitrogen	EPA 300.0	1	09182196601A	07/01/2009 12:43	Ashley M Adams	5
01506	Nitrite Nitrogen	EPA 300.0	1	09182196601A	07/01/2009 12:43	Ashley M Adams	5
00228	Sulfate	EPA 300.0	1	09182196601A	07/01/2009 12:43	Ashley M Adams	5
^226	Ortho-Phosphate as P	EPA 365.3	1	09182022601A	07/01/2009 00:30	Daniel S Smith	1

Approved
21 Jul 2009

*This limit was used in the evaluation of the final result

EXACT COPY
HPV 22 APR 2010

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax: 717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. WW 5712407

Group No. 1151471
PA

Sample #1 905 Analytical Grab Water Sample
Semi-Annual

Collected: 06/30/2009 10:11 by JF

Account Number: 02423

Submitted: 06/30/2009 16:30
Reported: 07/13/2009 at 14:33
Discard: 07/28/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

10905

CAT No.	Analysis Name		CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Dilution Factor
SW-846 8151A		Herbicides		ug/l	ug/l	ug/l	
01856	2,4-D		94-75-7	N.D.	0.16	0.49	1
01856	Dalapon		75-99-0	N.D.	0.25	1.2	1
01856	2,4-DB		94-82-6	N.D.	0.30	0.98	1
01856	Dicamba		1918-00-9	N.D.	0.079	0.30	1
01856	Dinoseb		88-85-7	N.D.	0.098	0.49	1
01856	2,4-DP (Dichlorprop)		120-36-5	N.D.	0.16	0.49	1
01856	MCPA		94-74-6	N.D.	300	980	1
01856	MCPP		93-65-2	N.D.	49	200	1
01856	Pentachlorophenol		87-86-5	N.D.	0.027	0.049	1
01856	2,4,5-T		93-76-5	N.D.	0.015	0.049	1
01856	2,4,5-TP		93-72-1	0.024 J	0.0098	0.049	1
EPA 608		Pesticides/PCBs		ug/l	ug/l	ug/l	
00178	Aldrin		309-00-2	N.D.	0.0041	0.019	1
00178	Alpha BHC		319-84-6	N.D.	0.0026	0.0097	1
00178	Beta BHC		319-85-7	N.D.	0.018	0.058	1
00178	Gamma BHC - Lindane		58-89-9	N.D.	0.0044	0.0097	1
00178	Chlordane		57-74-9	N.D.	0.068	0.48	1
00178	p,p-DDD		72-54-8	N.D.	0.0039	0.019	1
00178	p,p-DDE		72-55-9	N.D.	0.0048	0.019	1
00178	p,p-DDT		50-29-3	N.D.	0.0058	0.019	1
00178	Delta BHC		319-86-8	N.D.	0.0041	0.0097	1
00178	Dieldrin		60-57-1	N.D.	0.0039	0.019	1
00178	Endosulfan I		959-98-8	N.D.	0.0029	0.0097	1
00178	Endosulfan II		33213-65-9	N.D.	0.0039	0.019	1
00178	Endosulfan Sulfate		1031-07-8	N.D.	0.0039	0.019	1
00178	Endrin		72-20-8	N.D.	0.0039	0.019	1
00178	Endrin Aldehyde		7421-93-4	N.D.	0.019	0.097	1
00178	Heptachlor		76-44-8	N.D.	0.0039	0.0097	1
00178	Heptachlor Epoxide		1024-57-3	N.D.	0.0029	0.0097	1
00178	PCB-1016		12674-11-2	N.D.	0.097	0.48	1
00178	PCB-1221		11104-28-2	N.D.	0.15	0.48	1
00178	PCB-1232		11141-16-5	N.D.	0.097	0.48	1
00178	PCB-1242		53469-21-9	N.D.	0.097	0.48	1
00178	PCB-1248		12672-29-6	N.D.	0.097	0.48	1
00178	PCB-1254		11097-69-1	N.D.	0.097	0.48	1
00178	PCB-1260		11096-82-5	N.D.	0.097	0.48	1
00178	Toxaphene		8001-35-2	N.D.	0.29	0.97	1
EPA 200.7 rev 4.4		Metals		mg/l	mg/l	mg/l	
07035	Arsenic		7440-38-2	N.D.	0.0072	0.0200	1
07046	Barium		7440-39-3	N.D.	0.00060	0.0050	1
07049	Cadmium		7440-43-9	N.D.	0.0020	0.0050	1
07051	Chromium		7440-47-3	N.D.	0.0034	0.0150	1
07055	Lead		7439-92-1	N.D.	0.0069	0.0150	1
07036	Selenium		7782-49-2	N.D.	0.0089	0.0200	1
07066	Silver		7440-22-4	N.D.	0.0023	0.0050	1

*=This limit was used in the evaluation of the final result

Approved 21 Jul 2009

EXACT COPY
MPV 22 APR 2010

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax:717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. WW 5712407 **Group No. 1151471**
PA

Sample #1 905 Analytical Grab Water Sample
Semi-Annual

Collected: 06/30/2009 10:11 by JF

Account Number: 02423

Submitted: 06/30/2009 16:30
Reported: 07/13/2009 at 14:33
Discard: 07/28/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

10905

CAT No.	Analysis Name	CAS Number	As Received Result		As Received Method Detection Limit*	As Received Limit of Quantitation	Dilution Factor
EPA 200.7 rev 4.4	**Metals**		mg/l		mg/l	mg/l	
07072	Zinc	7440-66-6	N.D.		0.0081	0.0200	1
EPA 245.1 rev 3	**Metals**		mg/l		mg/l	mg/l	
00259	Mercury	7439-97-6	N.D.		0.000056	0.00020	1
EPA 300.0	**Wet Chemistry**		mg/l		mg/l	mg/l	
01505	Bromide	24959-67-9	N.D.		2.0	2.5	5
224	Chloride	16887-00-6	2.8		1.0	2.0	5
504	Fluoride	16984-48-8	1.1		0.40	0.50	5
00368	Nitrate Nitrogen	14797-55-8	N.D.		0.25	0.50	5
01506	Nitrite Nitrogen	14797-65-0	N.D.		0.40	0.50	5
00228	Sulfate	14808-79-8	2.7	J	1.5	5.0	5
EPA 365.3	**Wet Chemistry**		mg/l		mg/l	mg/l	
00226	Ortho-Phosphate as P	7723-14-0	N.D.		0.010	0.030	1

General Sample Comments

PA DEP Lab Certification ID 36-00037, Expiration Date: 1/31/10

All QC is compliant unless otherwise noted. Please refer to the Quality Control Summary for overall QC performance data and associated samples.

Laboratory Sample Analysis Record

CAT No.	Analysis Name	Method	Trial#	Batch#	Analysis Date and Time	Analyst	Dilution Factor
01856	Herbicides in Water	SW-846 8151A	1	091820017A	07/06/2009 16:51	John W Perkins	1
00178	Pesticides/PCB's in Water	EPA 608	1	091820005A	07/09/2009 02:20	Mark E McNulty	1
10241	Method 608 Water Extraction	EPA 608	1	091820005A	07/01/2009 17:00	JoElla L Rice	1
00816	Water Sample Herbicide Extract	SW-846 8151A	1	091820017A	07/02/2009 01:00	Tracy L Schickel	1
07035	Arsenic	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
07046	Barium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
07049	Cadmium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
07051	Chromium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
07055	Lead	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
07036	Selenium	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 19:00	John P Hook	1
07066	Silver	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
07072	Zinc	EPA 200.7 rev 4.4	1	091875716006	07/09/2009 01:59	Tara L Snyder	1
^359	Mercury	EPA 245.1 rev 3	1	091875714003	07/09/2009 17:28	Parker D Lindstrom	1
/16	EPA 600 ICP Digest (tot rec)	EPA 200.7 rev 4.4	1	091875716006	07/08/2009 14:26	James L Mertz	1

*=This limit was used in the evaluation of the final result

Approved MD Allen 21-Jul2009

EXACT COPY
HPJ 22 APR 2010

Lancaster Laboratories

Analysis Report

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax:717-656-2681 • www.lancasterlabs.com

Lancaster Laboratories Sample No. WW 5712407

Group No. 1151471
PA

Sample #1 905 Analytical Grab Water Sample
Semi-Annual

Collected: 06/30/2009 10:11 by JF

Account Number: 02423

Submitted: 06/30/2009 16:30
Reported: 07/13/2009 at 14:33
Discard: 07/28/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

10905

Laboratory Sample Analysis Record

CAT No.	Analysis Name	Method	Trial#	Batch#	Analysis Date and Time	Analyst	Dilution Factor
05714	PW/WW Hg Digest	EPA 245.1 rev 3	1	091875714003	07/08/2009 15:30	James L Mertz	1
01505	Bromide	EPA 300.0	1	09182196601A	07/01/2009 14:05	Ashley M Adams	5
00224	Chloride	EPA 300.0	1	09182196601A	07/01/2009 14:05	Ashley M Adams	5
01504	Fluoride	EPA 300.0	1	09182196601A	07/01/2009 14:05	Ashley M Adams	5
00368	Nitrate Nitrogen	EPA 300.0	1	09182196601A	07/01/2009 14:05	Ashley M Adams	5
01506	Nitrite Nitrogen	EPA 300.0	1	09182196601A	07/01/2009 14:05	Ashley M Adams	5
00228	Sulfate	EPA 300.0	1	09182196601A	07/01/2009 14:05	Ashley M Adams	5
[illegible]226	Ortho-Phosphate as P	EPA 365.3	1	09182022601A	07/01/2009 00:30	Daniel S Smith	1

Approved
[signature]
21-Jul 2009

*=This limit was used in the evaluation of the final result

EXACT COPY
HPV 221 APR 2010

BEDDING ANALYSIS

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax:717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. G5 5764007

Bedding Sample Lot# 082409

Group No. 1159797
PA

Collected: 08/27/2009

Account Number: 02423

Submitted: 08/28/2009 17:00
Reported: 09/09/2009 at 07:03
Discard: 09/24/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

82409

CAT No.	Analysis Name	CAS Number	As Received Result	As Received Method Detection Limit*	As Received Limit of Quantitation	Dilution Factor
Herbicides	**SW-846 8151A**		ug/kg	ug/kg	ug/kg	
01863	2,4-D	94-75-7	15 J	12	36	1
01863	2,4,5-TP	93-72-1	N.D.	0.75	1.7	1
Pesticides/PCBs	**SW-846 8081A**		ug/kg	ug/kg	ug/kg	
06005	Aldrin	309-00-2	N.D.	0.33	0.83	1
06005	Alpha BHC	319-84-6	N.D.	0.17	0.83	1
06005	Beta BHC	319-85-7	N.D.	0.19	0.83	1
005	Gamma BHC - Lindane	58-89-9	N.D.	0.17	0.83	1
005	Chlordane	57-74-9	N.D.	4.0	17	1
06005	p,p-DDD	72-54-8	N.D.	0.33	1.7	1
06005	p,p-DDE	72-55-9	N.D.	0.33	1.7	1
06005	p,p-DDT	50-29-3	N.D.	0.33	1.7	1
06005	Delta BHC	319-86-8	N.D.	0.31	0.83	1
06005	Dieldrin	60-57-1	N.D.	0.33	1.7	1
06005	Endosulfan I	959-98-8	N.D.	0.22	0.83	1
06005	Endosulfan II	33213-65-9	N.D.	0.33	1.7	1
06005	Endosulfan Sulfate	1031-07-8	N.D.	0.33	1.7	1
06005	Endrin	72-20-8	N.D.	0.33	1.7	1
06005	Endrin Aldehyde	7421-93-4	N.D.	0.33	1.7	1
06005	Heptachlor	76-44-8	N.D.	0.17	0.83	1
06005	Heptachlor Epoxide	1024-57-3	N.D.	0.17	0.83	1
06005	Methoxychlor	72-43-5	N.D.	1.7	8.3	1
06005	Toxaphene	8001-35-2	N.D.	11	33	1
Pesticides/PCBs	**SW-846 8082**		ug/kg	ug/kg	ug/kg	
02033	PCB-1016	12674-11-2	N.D.	3.30	17.0	1
02033	PCB-1221	11104-28-2	N.D.	3.30	17.0	1
02033	PCB-1232	11141-16-5	N.D.	3.30	17.0	1
02033	PCB-1242	53469-21-9	N.D.	3.30	17.0	1
02033	PCB-1248	12672-29-6	N.D.	3.30	17.0	1
02033	PCB-1254	11097-69-1	N.D.	3.30	17.0	1
02033	PCB-1260	11096-82-5	N.D.	3.30	17.0	1
Metals	**SW-846 6010B**		mg/kg	mg/kg	mg/kg	
05935	Arsenic	7440-38-2	N.D.	0.941	1.98	1
05946	Barium	7440-39-3	0.666	0.0396	0.495	1
05949	Cadmium	7440-43-9	N.D.	0.139	0.495	1
05951	Chromium	7440-47-3	N.D.	0.584	1.49	1
05955	Lead	7439-92-1	N.D.	0.594	1.49	1
05936	Selenium	7782-49-2	N.D.	0.970	1.98	1
06966	Silver	7440-22-4	N.D.	0.178	0.495	1
	SW-846 7471A		mg/kg	mg/kg	mg/kg	
159	Mercury	7439-97-6	N.D.	0.0114	0.0995	1

*=This limit was used in the evaluation of the final result

Approved
21 sep 2009

EXACT COPY
HPV 22 APR 2010

Lancaster Laboratories

2425 New Holland Pike, PO Box 12425, Lancaster, PA 17605-2425 • 717-656-2300 Fax:717-656-2681 • www.lancasterlabs.com

Analysis Report

Lancaster Laboratories Sample No. G5 5764007

Group No. 1159797
PA

Bedding Sample Lot# 082409

Collected: 08/27/2009

Account Number: 02423

Submitted: 08/28/2009 17:00
Reported: 09/09/2009 at 07:03
Discard: 09/24/2009

Charles River Laboratories
905 Sheehy Dr.
Horsham PA 19044-1297

82409

General Sample Comments

PA DEP Lab Certification ID 36-00037, Expiration Date: 1/31/10

All QC is compliant unless otherwise noted. Please refer to the Quality Control Summary for overall QC performance data and associated samples.

Laboratory Sample Analysis Record

CAT No.	Analysis Name	Method	Trial#	Batch#	Analysis Date and Time	Analyst	Dilution Factor
963	Appendix IX Herbicides in Soil	SW-846 8151A	1	092440011A	09/05/2009 08:21	Michele D Hamilton	1
005	Pesticides in Solids	SW-846 8081A	1	092430010A	09/03/2009 14:45	Lindsey K Lafferty	1
02033	PCBs in Soil	SW-846 8082	1	092430011A	09/03/2009 12:26	Jamie L Brillhart	1
06006	PPL Pesticide Solid Extraction	SW-846 3550B	1	092430011A	08/31/2009 21:15	Karen L Beyer	1
06006	PPL Pesticide Solid Extraction	SW-846 3550B	2	092430010A	08/31/2009 21:15	Karen L Beyer	1
04181	Herbicide Soil Extraction	SW-846 3550B/SW-846 8151A	1	092440011A	09/01/2009 20:25	Karen L Beyer	1
06935	Arsenic	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
06946	Barium	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
06949	Cadmium	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
06951	Chromium	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
06955	Lead	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
06936	Selenium	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
06966	Silver	SW-846 6010B	1	092445708004	09/02/2009 18:06	John P Hook	1
00159	Mercury	SW-846 7471A	1	092445711001	09/02/2009 19:44	Nelli S Markaryan	1
05708	SW SW846 ICP Digest	SW-846 3050B	1	092445708004	09/01/2009 19:55	Annamaria Stipkovits	1
05711	SW SW846 Hg Digest	SW-846 7471A modified	1	092445711001	09/01/2009 23:55	Annamaria Stipkovits	1

Approved
21 sep 2009

*=This limit was used in the evaluation of the final result

EXACT COPY
nw221 APR 12009