Laboratory Report

Analytical Report of Data for PFOS Dietary LC50 Study with Mallards (Sera and Liver)

Laboratory Report No. FACT-TOX-130 (W1902)

Attachment A

Sera Table of Results
Day 8
Day 22

Testing Laboratory

3M Environmental Technology & Safety Services
3M Environmental Laboratory
Fluorine Analytical Chemistry Team (FACT)
2-3E-09
935 Bush Avenue,
St. Paul, MN 55106

FACT-TOX-130

Study:	PFOS Dietary LC50 Study with Mallards
Product Number(Test Substance):	PFOS
Matrix:	Mallard Serum
Method/Revision:	ETS-8-4.1 and ETS-8-5.1
Analytical Equipment System Number:	Soup 020199, Amelia 062498
Instrument Software/Version:	Masslynx 3.2
Filename:	See Attachments
R-Squared Value:	See Attachments
Slope:	See Attachments
Y-Intercept:	See Attachments
Dates of Extraction/Analyst:	06/29/99 MCH
Dates of Analysis/Analyst:	07/01/99, 07/06/99, 07/08/99 DRB/MEE
Date of Data Reduction/Analyst:	07/02/99, 0707/99, 07/09/99 DRB/MEE

Sample Data

MALLARD SERUM Day 8

Group Dose	Sample #	[PFOS] Reported (ug/mL) or %Rec	Average [PFOS]ug/mL or %Rec	RSD(%) Std. Dev.(ug/mL)
Method Blk	H2O Blk-1	<LOQ		
	H2O Blk-2	<LOQ		
	H2O Blk-3	<LOQ		
	H2O Blk-4	<LOQ	<LOQ	NA
Matrix Blk	Rabbit Serum Blk-1	0.00742		
	Rabbit Serum Blk-2	<LOQ		
	Rabbit Serum Blk-3	0.00730		16.4
	Rabbit Serum Blk-4	0.00967	0.00813 - 1 Outlier	0.00133
QC - 50 ppb=MS	RBS06299-MS-1	107%		
1.0 ppm = MSD	RBS06299-MSD-1	74%		
	RBS06299-MS-2	103%	50 ppb	
	RBS06299-MSD-2	78%	101%	5%
	RBS06299-MS-3	96%		
	RBS06299-MSD-3	126%		
	RBS06299-MS-4	99%	1.0 ppm	
	RBS06299-MSD-4	119%	100%	27%
Group1	5938	<LOQ		
0.0 mg/kg	5939	<LOQ		
	5940	<LOQ		
	5941	<LOQ		
	5942	<LOQ		
	9195	<LOQ		
	9196	0.0103		
	9197	0.00686		
	9198	<LOQ		
	9199	<LOQ		
	9200	<LOQ		
	8401	0.00674		
	8402	<LOQ		
	8403	0.00819		
	8404	0.0230	<LOQ - 5 Outliers	NA
Group2	9190	8.81		
9.10 mg/kg food	9191	7.68		
	9192	8.79		
	9193	4.04		26.5
	9194	7.60	7.38	1.96
Group3	5958	9.31		
18.3 mg/kg food	5959	25.6		
	5960	11.45		
	5961	8.89		49.8
	5962	13.7	13.8	6.86
Group4	5968	27.3		
36.6 mg/kg food	5969	29.2		
	5970	17.7		
	5971	44.3		31.9
	5972	34.2	30.5	9.75
Group5	5988	37.8		
73.2 mg/kg food	5989	41.6		
	5990	51.3		
	5991	57.8		17.0
	5992	52.0	48.1	8.19
Group6	5998	63.6		
146 mg/kg food	5999	50.6		
	6000	35.7		
	9173	81.2		34.8
	9174	38.6	53.9	18.8
Group7	9180	67.2		
293 mg/kg food	9181	58.9		9.57
	9183	71.2	65.8	6.29
Group8	5978	68.4		
586 mg/kg food	5979	94.5		43.4
	5981	159	107	46.5
Group 9	NS	NA	NA	NA
1171 mg/kg food				

Limit of Quantitation (LOQ) PFOS = 0.005 ug/mL

PFOS = Perfluorooctanesulfonate

Extraction volume ratio equals initial volume/final volume.

Date Entered/By: 7/06/99, 7/07/99, 07/14/99 LAC

Date Verified/ By: 08/09/99 GML

NA = not applicable

NS = no sample analyzed

FACT-TOX-130

Study:	PFOS Dietary LC50 Study with Mallards
Product Number(Test Substance):	PFOS
Matrix:	Mallard Serum
Method/Revision:	ETS-8-4.1 and ETS-8-5.1
Analytical Equipment System Number:	Soup 020199
Instrument Software/Version:	Masslynx 3.2
Filename:	See Attachments
R-Squared Value:	See Attachments
Slope:	See Attachments
Y-Intercept:	See Attachments
Dates of Extraction/Analyst:	07/07/99 RWW
Dates of Analysis/Analyst:	07/12/99, 07/15/99 DRB/GML
Date of Data Reduction/Analyst:	07/13/99, 07/16/99 DRB/KJH

Sample Data

MALLARD SERUM Day 22

Group Dose	Sample #	[PFOS] Reported (ug/mL) or %Rec	Average [PFOS]ug/mL or %Rec	RSD(%) Std. Dev.(ug/mL)
Method Blk	H2O Blk-1	<LOQ		
	H2O Blk-2	<LOQ		
	H2O Blk-3	<LOQ		
	H2O Blk-4	<LOQ	<LOQ	NA
Matrix Blk	Rabbit Serum Blk-1	<LOQ		
	Rabbit Serum Blk-2	<LOQ		
	Rabbit Serum Blk-3	<LOQ		
	Rabbit Serum Blk-4	<LOQ	<LOQ	NA
QC - 50 ppb	RBS07079-MS-1	104%		
	RBS07079-MSD-1	100%		
	RBS07079-MS-2	103%		
	RBS07079-MSD-2	99%	101%	2%
1000 ppb	RBS07079-MS-1	99%		
	RBS07079-MSD-1	97%		
	RBS07079-MS-2	107%		
	RBS07079-MSD-2	95%	100%	6%
Group1	6495	<LOQ		
0.0 mg/kg food	6496	<LOQ		
	6497	<LOQ		
	6498	<LOQ		
	6499	<LOQ		
	6500	<LOQ		
	5929	<LOQ		
	5930	<LOQ		
	5931	<LOQ		
	5932	<LOQ		
	5933	<LOQ		
	5934	<LOQ		
	5935	<LOQ		
	5936	<LOQ		NA
	5937	<LOQ	<LOQ	NA
Group2	9185	1.05		
9.10 mg/kg food	9186	1.09		
	9187	2.68		
	9188	1.04		47.3
	9189	1.58	1.49	0.704
Group3	5953	4.40		
18.3 mg/kg food	5954	3.81		
	5955	4.70		
	5956	4.92		12.0
	5957	3.75	4.32	0.520
Group4	5963	4.30		
36.6 mg/kg food	5964	5.83		
	5965	7.27		
	5966	7.01		29.1
	5967	9.69	6.82	1.99
Group5	5983	24.5		
73.2 mg/kg food	5984	10.0		
	8985	6.52		
	8986	7.23		66.1
	8987	8.43	11.3	7.49
Group6	5993	12.2		
146 mg/kg food	5994	11.9		
	5995	16.2		
	5996	24.0		42.0
	5997	30.1	18.9	7.94
Group7	9175	10.3		
293 mg/kg food	9176	8.43		
	9177	10.1		
	9178	16.5		41.2
	9179	21.5	13.4	5.51
Group8	5943	29.7		
586 mg/kg food	5944	15.2		
	5945	39.4		45.3
	5947	51.6	34.0	15.4
*Group9	5943	25.5	NA	NA
1171 mg/kg food				

Limit of Quantitation (LOQ) PFOS = 0.010 ug/mL

PFOS = Perfluorooctanesulfonate

NA = not applicable

*Grp 9 tentative value, evaporated to dryness.

Extraction volume ratio equals initial volume/final volume.

Date Entered/By: 07/14/99, 07/16/99, 07/20/99 LAC

Date Verified/ By: 08/09/99 GML

Laboratory Report

Analytical Report of Data for PFOS Dietary LC50 Study with Mallards (Sera and Liver)

Laboratory Report No. FACT-TOX-130 (W1902)

Attachment B
Liver Table of Results
Day 4-7
Day 8
Day 22

Testing Laboratory

3M Environmental Technology & Safety Services
3M Environmental Laboratory
Fluorine Analytical Chemistry Team (FACT)
2-3E-09
935 Bush Avenue,
St. Paul, MN 55106

Study: PFOS Dietary LC50 Study with Mallards
Product Number(Test Substance): PFOS
Matrix: Mallard Liver

MALLARD LIVER DAY 4-7

Group Dose	Sample #	PFOS Calc. Conc. ng/g	Concentraion of PFOS ug/g or % Rec.	Mean PFOS ug/g or % Rec.	RSD (%) Std. Dev.(ug/g) MS/MSD RPD
Method Blk	MLL09279-H2O Blk-11	0.00	<LOQ		
	MLL09279-H2O Blk-12	0.00	<LOQ	<LOQ	NA
QC - 50 ppb**	MLL09279-9196-0-MS-11	31.1	56%		
	MLL09279-9196-0-MSD-11	182	325%	191%	141%
QC - 1.0 ppm**	MLL09279-9199-0-MS-12	677	61%		
	MLL09279-9199-0-MSD-12	536	49%	55%	23%
Group 7 293 mg/kg food	9182, Day7	148106	148	198	70.7
Group8	5980, Day5	216457	216		
586 mg/kg food	5982, Day6	196822	197		9.23
	5946, Day7	179996	180	198	18.2
Group9	5976, Day4	115890	116		
1171 mg/kg food	5949, Day4	122122	122		
	5952, Day5	172280	172		
	5950, Day5	137062	137		
	5974, Day5	146732	147		
	5951, Day5	185296	185		
	5973, Day6	131286	131		
	5948, Day6	120973	121		18.3
	5975, Day7	177851	178	145	26.6

PFOS = Perfluorooctanesulfonate
Limit of Quantitation (LOQ) PFOS: approx. 0.030ug/g
NA = not applicable
**The MS/MSD and group 1 samples were re-extracted on 10/08/99 to confirm matrix effects. LAC 10/08/99

FACT-TOX-130

Study: PFOS Dietary LC50 Study with Mallards
Product Number(Test Substance): PFOS
Matrix: Mallard Liver

MALLARD LIVER DAY 8

Group Dose	Sample #	PFOS Calc. Conc. ng/g	Concentration of PFOS ug/g or % Rec.	Mean PFOS ug/g or % Rec.	RSD (%) Std. Dev.(ug/g) MS/MSD RPD
Method Blk	H2O Blk-1	0.00	<LOQ		
	H2O Blk-2	0.00	<LOQ		
	H2O Blk-3	0.00	<LOQ		
	H2O Blk-4	0.00	<LOQ	<LOQ	NA
QC - 100 ppb	MLL06299-5938-0-MS-1	88.0	74%		
	MLL06299-5938-0-MSD-1	89.4	75%		
1.0 ppm	MLL06299-5940-0-MS-2	802	73%	100 ppb	
	MLL06299-5940-0-MSD-2	799	72%	73%	5%
100 ppb	MLL06299-9196-0-MS-3	84.4	76%		
	MLL06299-9196-0-MSD-3	73.3	66%		
1.0 ppm	MLL06299-8401-0-MS-4	915	76%	1.0 ppm	
	MLL06299-8401-0-MSD-4	894	75%	74%	2%
Group1	5938	0.00	<LOQ		
0.0 mg/kg food	5939	0.00	<LOQ		
	5940	0.00	<LOQ		
	5941	0.00	<LOQ		
	5942	0.00	<LOQ		
	9195	0.00	<LOQ		
	9196	92.8	0.0928		
	9197	0.00	<LOQ		
	9198	0.00	<LOQ		
	9199	132	0.132		
	9200	0.00	<LOQ		
	8401	0.00	<LOQ		
	8402	0.00	<LOQ		
	8403	0.00	<LOQ		NA
	8404	0.00	<LOQ	<LOQ - 2 Outliers	NA
Group2	9190	6828	6.83		
9.10 mg/kg food	9191	3714	3.71		
	9192	4690	4.69		
	9193	2902	2.90		32.1
	9194	5217	5.22	4.67	1.50
Group3	5958	4760	4.76		
18.3 mg/kg food	5959	11542	11.5		
	5960	6158	6.16		
	5961	4202	4.20		43.7
	5962	6562	6.56	6.65	2.90
Group4	5968	14690	14.7		
36.6 mg/kg food	5969	14901	14.9		
	5970	9864	9.86		
	5971	20377	20.4		24.8
	5972	16505	16.5	15.3	3.78
Group5	5988	15347	15.3		
73.2 mg/kg food	5989	16890	16.9		
	5990	29729	29.7		
	5991	51544	51.5		49.8
	5992	34849	34.8	29.7	14.78
Group6	5998	38069	38.1		
146 mg/kg food	5999	35398	35.4		
	6000	32775	32.8		
	9173	55034	55.0		28.1
	9174	32084	32.1	38.8	10.9
Group7	9180	59768	59.8		
293 mg/kg food	9181	56336	56.3		
	9183	50650	50.6		8.29
	9184*	248075	248	55.6	4.61
Group8	5978	32469	32.5		
586 mg/kg food	5979	75542	75.5		39.6
	5981	70474	70.5	59.5	23.5
Group9-1171 mg/kg food	NS	NA	NA	NA	NA

PFOS = Perfluorooctanesulfonate
Limit of Quantitation (LOQ) PFOS: approx. 0.030ug/g
NA = not applicable
NS = no sample analyzed
* One outlier, not included in any calculations

FACT-TOX-130

Study: PFOS Dietary LC50 Study with Mallards
Product Number(Test Substance): PFOS
Matrix: Mallard Liver

MALLARD LIVER DAY 22

Group Dose	Sample #	PFOS Calc. Conc. ng/g	Concentration of PFOS ug/g or % Rec.	Mean PFOS ug/g or % Rec.	RSD (%) Std. Dev.(ug/g) MS/MSD RPD
Method Blk	H2O Blk-1	1.72	<LOQ		
	H2O Blk-2	1.09	<LOQ	<LOQ	NA
	H2O Blk-3	1.07	<LOQ		
	H2O Blk-4	1.51	<LOQ	<LOQ	NA
QC - 100 ppb	6495-0-MS-1	107	90%		
	6495-0-MSD-1	101	85%		
	6496-0-MS-2	111	94%		
	6496-0-MSD-2	126	107%	94%	9%
1.0 ppm	6497-0-MS-3	1033	87%		
	6497-0-MSD-3	1111	93%		
	6498-0-MS-4	1093	91%		
	6498-0-MSD-4	1053	88%	90%	3%
Group1	6495	22.2	<LOQ		
0.0 mg/kg food	6496	19.3	<LOQ		
	9497	12.8	<LOQ		
	6498	13.2	<LOQ		
	6499	8.53	<LOQ		
	6500	13.4	<LOQ		
	5929	5.63	<LOQ		
	5930	4.51	<LOQ		
	5931	1.72	<LOQ		
	5932	3.10	<LOQ		
	5933	8.99	<LOQ		
	5934	8.49	<LOQ		
	5935	9.36	<LOQ		
	5936	4.83	<LOQ		NA
	5937	5.51	<LOQ	<LOQ	NA
Group2	9185	911	0.911		
9.10 mg/kg food	9186	876	0.876		
	9187	1569	1.57		
	9188	735	0.735		31.4
	9189	1038	1.04	1.03	0.322
Group3	5953	1802	1.80		
18.3 mg/kg food	5954	2186	2.19		
	5955	1930	1.93		
	5956	2667	2.67		20.3
	5957	1594	1.59	2.04	0.413
Group4	5963	1626	1.63		
36.6 mg/kg food	5964	3035	3.03		
	5965	3154	3.15		
	5966	3639	3.64		40.0
	5967	5366	5.37	3.36	1.35
Group5	5983	10254	10.3		
73.2 mg/kg food	5984	8812	8.81		
	5985	7957	7.96		
	5986	10847	10.8		13.9
	5987	8195	8.20	9.21	1.28
Group6	5993	13976	14.0		
146 mg/kg food	5994	7737	7.74		
	5995	8374	8.37		
	5996	14015	14.0		38.1
	5997	19574	19.6	12.7	4.85
Group7	9175	7494	7.49		
293 mg/kg food	9176	5633	5.63		
	9177	7687	7.69		
	9178	12522	12.5		34.8
	9179	12663	12.7	9.20	3.20
Group8	5943	18754	18.8		
586 mg/kg food	5944	9763	9.76		
	5945	24677	24.7		38.8
	5947	27443	27.4	20.2	7.82
Group9-1171 mg/kg food	5977	16261	16.3	NA	NA

PFOS = Perfluorooctanesulfonate
Limit of Quantitation (LOQ) PFOS: approx. 0.030ug/g
NA = not applicable

Laboratory Report – Revision 1

Analytical Report of Data for PFOS Dietary LC50 Study with Mallards (Sera and Liver)

Laboratory Report No. FACT-TOX-130 (W1902)

Testing Laboratory

3M Environmental Technology & Safety Services
3M Environmental Laboratory
Fluorine Analytical Chemistry Team (FACT)
2-3E-09
935 Bush Avenue,
St. Paul, MN 55106

Laboratory Contact

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Revision Date: March 19, 2001

1 Introduction

This study was designed to quantitatively assess perfluorooctanesulfonate (PFOS, C8F17SO3-, CAS # 2795-39-3) levels in the sera and liver of mallard ducks exposed to PFOS through diet. Details of dosing conditions, clinical observations and other toxicity data were generated by the in-life testing facility (Wildlife International) and are not included in this report. Subsets of the nine groups of test animals were sacrificed on day 8 and day 22, post-dose. Sera and liver collected from the sacrificed animals were sent to the 3M Environmental Lab and analyzed quantitatively for PFOS. Tissues from some animals that died before the scheduled sacrifice date were collected and analyzed also; results are included in this report.

With few exceptions, PFOS levels in the sera and livers of the mallards increased with dose group. Additionally, considering the average PFOS levels within each dose group, both liver and sera levels of PFOS declined dramatically between day 8 and day 22. In most animals, for a specific time point, the ratio of PFOS concentration in sera (μg/mL) to PFOS concentration in liver (μg/g) was greater than 1.

No data was collected for analytes other than PFOS.

Although rigorous quality control measures and 3M Environmental Laboratory Standard Operating Procedures were followed, the acquisition of this data was not necessarily collected according to applicable Good Laboratory Practices. Data presented here is the highest quality data available at this time.

In-phase testing was conducted according to applicable Good Laboratory Practices. The conduct of the in-life phase is the sole responsibility of Wildlife International Ltd. See attachment C for the in-life protocol.

2 Sample Receipt

The Environmental Lab received 60 liver, 58 bile, 46 blood, and 46 serum specimens, frozen, on 05/19/99. Upon specimen receipt, a 3M chain of custody was initiated.

The Environmental Lab received 50 liver, 50 bile, 50 blood, and 50 serum samples, frozen, on 06/29/99. Upon specimen receipt, a 3M chain of custody was initiated. A copy of each form can be found in an Attachment to this report.

3 Holding Times

There is no holding time criteria associated with these samples for LC/ESMSMS analysis.

4 Methods - Analytical and Preparatory

Analytical methods

ETS-8-4.1, Extraction of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds from Serum for Analysis Using HPLC-Electrospray/Mass Spectrometry *with the following exceptions*:
Due to the small volume of mallard sera available, standard curves were prepared in methanol instead of from mallard sera. Available sample volumes were so small, there was not enough sera for matrix spike studies. As an indicator of gross error, rabbit serum was used as a surrogate matrix. Sixteen matrix spikes were prepared in rabbit sera. PFOS lot 171 was used as reference material.
ETS-8-6.0, Extraction of Potassium Perfluorooctanesulfonate or Other Fluorochemical Compounds from Liver for Analysis Using HPLC-Electrospray/Mass Spectrometry *with the following exception*:

Due to the small amount of mallard liver available, standard curves were prepared in methanol instead of from mallard liver. Twelve matrix spikes were prepared from the mallard liver. As described in the method, all results are reported in terms of µg of analyte per gram of liver (wet weight). PFOS lot 171 was used as reference material.

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

ETS-8-7.0, Analysis of Potassium Perfluorooctanesulfonate or Other Fluorochemical in Liver Extracts Using HPLC-Electrospray/Mass Spectrometry

Sample preparation –HPLC/ESMSMS: ion-pairing extraction

Analyte is extracted from a sample matrix with ion pairing reagent [tetrabutyl ammonium hydrogen sulfate (TBA)] in a pH-controlled environment. The cationic reagent selectively targets anionic fluorochemicals. Once the anion-TBA pair is formed, the analyte is transferred into a non-polar organic solvent (methyl-*tert*-butyl ether), dried, and reconstituted in methanol for MS analysis.

HPLC/ESMSMS

In HPLC, an aliquot of extract is injected and passed through a reverse phase liquid chromatographic column. Based on the affinity of the analyte for the stationary phase in the column relative to the liquid mobile phase, the analyte is retained for a characteristic amount of time. For example, in a standard solution PFOS may elute at 8.0 minutes. Retention times between a standard PFOS solution and the analyte extracted from tissue in this analysis were matched on the HPLC system to within 1%.

Following HPLC separation, ESMSMS provides a rapid and accurate means for analyzing a wide range of organic compounds, including fluorochemicals. Electrospray, an ionization technique used primarily for detection of molecular ions, is generally operated at relatively mild temperatures. Molecules are ionized, and a primary ion, characteristic of the analyte, is selected. Ions are accelerated for collision with argon gas resulting in fragmentation of the selected species; subsequent collisions create smaller secondary ionic fragments unique to the primary ion, which are detected.

For example, for PFOS ($C_8F_{17}SO_3^-$) analysis, ion 499 is selected as the characteristic primary ion. This ion is fragmented into other ions such as 80 amu (corresponding to SO_3^-), and 230 amu ($C_3F_6SO_3^-$). Each of these secondary fragments is detected and can be used to differentiate PFOS from other compounds that might have the same characteristic 499 amu primary ion, but different chemical compositions and secondary ion fragmentation patterns.

5 Analysis

5.1 Calibration

For quantitative determinations, a mid-level, calibration curve verification (CCV) standard in methanol was analyzed every 5 to 10 samples to monitor instrumental drift. Calibration curve verification standards are acceptable if instrumental response is within +/- 30% of the expected value. In this study, all CCV standards were acceptable.

Quantitation of the target analytes is based on linear regression analysis (weighted 1/x) of two curves, prepared in methanol, bracketing each group of samples. Quantitation of each analyte is based on the response of one or more specific daughter ions using the multiple response-monitoring mode of the instrument.

3M lot #217 (purity 86.9%) of PFOS was used for calibration standards. As purity information was not available at the time of data collection, reported results have not been corrected.

5.2 Blanks

Two to four extraction blanks, utilizing water as surrogate matrix, were extracted with each batch of samples. Blanks were acceptable if no target analyte was detected above the limit of detection for a specific analyte. In this study, all blanks were acceptable.

5.3 Surrogates

1,1,2,2-Tetra-hydroperfluorooctanesulfonate was used as a surrogate in this analysis. Surrogate response was monitored to confirm gross instrumental failure, if necessary. Surrogate response was acceptable for all analyses.

5.4 Matrix Spikes

Over the course of the analysis, twenty-eight matrix spikes were prepared in the mallard liver matrix. With two exceptions, all matrix spikes showed acceptable recoveries of greater than 50%. Of the four matrix spikes extracted on 9/27/99, two were recovered outside the acceptable range of +/-50%. One sample had a PFOS recovery of 49% and a second sample showed PFOS recovery of 325%. These outlying recoveries were confirmed, but no obvious error was readily identified. An error in spiking the matrix is suspected. Because the remaining 26 matrix spikes were within the acceptable criteria, no further action was taken to explain these two anomalies. Finally, due to a miscalculation early in the data reduction process, eight additional matrix spikes were extracted to confirm what was thought to be low recovery. Upon discovery of the calculation error, the original matrix spikes were confirmed to be acceptable and the additional re-extractions were not used.

Because there was so little mallard sera available, rabbit sera was used as a surrogate matrix in this study. Sixteen matrix spikes were prepared in rabbit sera. All matrix spikes showed acceptable recovery of 70-130%.

5.5 Laboratory Control Samples

Laboratory Control Samples are not a component of this study.

5.6 Sample Related Comments

Samples were not analyzed for any residual or potential metabolite other than PFOS. Other matrices submitted with this study will be analyzed as needed upon written request from the sponsor. In the event of further analysis, an additional report will be generated.

6 Data Summary

With few exceptions, PFOS levels in the sera and livers of the mallards increased with dose group. Additionally, considering the average PFOS levels within each dose group, both liver and sera levels of PFOS declined dramatically between day 8 and day 22. Sera levels of PFOS measured in animals sacrificed on day 22 are between 20-35% of the sera levels of PFOS measured on day 8. Levels of PFOS in the livers of animals sacrificed on day 22 were, on average, 30% lower than those levels determined in the livers of animals sacrificed on day 8.

In most animals, for a specific time point, the ratio of PFOS concentration in sera (μg/mL) to PFOS concentration in liver (μg/g) was greater than 1.

7 Data Quality Objectives and Data Integrity

No circumstances existed during the present study that would have adversely affected the quality or integrity of the data. The following data quality objectives were followed during the study:

- Linearity – The coefficient of determination (r^2) of the standard curve was equal to or greater than 0.985 using 1/x weighting.
- Limits of quantitation - PFOS in sera is 0.005-0.010 µg/mL; PFOS in liver is approximately 0.030 µg / g
- Acceptable precision of duplicates - < 20%, determined by MS/MSD analysis
- Acceptable recoveries of spikes– 70% to 130% for sera; 50% to 150% for liver
- Use of confirmatory methods – Given the selectivity of the analytical tool used (HPLC-ESMSMS) and lack of a viable alternative for analysis, no confirmatory methods were used.
- Demonstration of specificity – Specificity was demonstrated by chromatographic retention time (matched to standards to within 3%) and the response of at least one characteristic product ion arising from collisions of an analyte-specific parent ion.
- The characterization of the reference material (PFOS, lot #171), with respect to purity, stability, and homogeneity, was not complete on the date this report was issued. Once the characterization is complete, a copy of the report will on file in the 3M Environmental Lab archives.

Given the parameters listed above, assuming spike recoveries form a suitable indication of endogenous analyte recovery, and assuming that rabbit sera is a suitable surrogate for mallard sera, sera data are quantitative to +/- 30%; liver data is quantitative to +/- 50%. The validity of these assumptions has not been verified by other techniques. If more accuracy is required, additional matrix should be supplied so samples can be evaluated versus a standard curve extracted from the same matrix as the samples.

8 Attachments

- Attachment A: Table of results of sera analysis
- Attachment B: Table of results of liver analysis
- Attachment C: In-life protocol
- Attachment D: Sample receipt
- Attachment E: Extraction and analytical data

9 Signatures

Mark Ellefson, Technologist — 03/23/01
Date

Kris Hansen, Ph.D., Principal Analytical Investigator — 03/23/01
Date

QAU review by Robert Voyksner
R.L. Kris 03/23/01

9 Signatures

Mark Ellefson, Technologist — Date: 06/05/00

Kris Hansen, Ph.D., Principal Analytical Investigator — Date: 06/06/00

QAU review by Robert Voyksner

Signature page from original report.

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