

AR226-1722

**Quantitative Determination of
PFOS, PFOSA, PFOSAA, PFHS,
POAA, M556 and M570 in Human
Serum by LC/MS/MS**

**Sample Analysis Report for Protocol
EPI-Swed**

**Northwest Bioanalytical (NWB)
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Salt Lake City, UT 84124**

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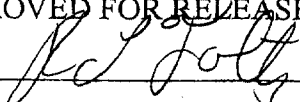


DATE:

12/8/99

David Vollmer, Ph.D., NWB Project Manager

APPROVED FOR RELEASE BY:



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12/9/99

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Northwest Bioanalytical

Study No. NWBS99-116
Report No. NWBR99-113

QUALITY ASSURANCE STATEMENT

LABORATORY: Northwest Bioanalytical (NWB)
A Division NWT, Inc.
1121 East 3900 South
Salt Lake City, UT 84124

SPONSOR: 3M Environmental Technology Services (3M)
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COMPOUND(S): PFOS, PFOSA, PFOSAA, PFHS, POAA, M556 and M570

NWB STUDY NUMBER: NWBS99-116

SPONSOR STUDY NUMBER: EPI-Swed

NWB STUDY TITLE: Extraction and Analysis of PFOS and other Fluorochemicals in
Human Serum by HPLC/Electrospray Mass Spectrometry

The clinical sample analysis study described in this report is not included within the definition of a GLP regulated nonclinical study. However, Northwest Bioanalytical conducts all studies within the guidelines of the U.S. FDA Good Laboratory Practice Regulations for Nonclinical Laboratory Studies (Title 21 CFR Part 58), the OECD Principles of Good Laboratory Practice and the Japanese MHW Good Laboratory Practice Standard Ordinance for Nonclinical Laboratory Studies on the Safety of Drugs (Ordinance No. 21, PAB Notification No. 424.) The following inspections were performed by the NWB QAU per SOP.

Inspection and Reporting Statement.

<u>Inspection Date</u>	<u>Phase of Study</u>	<u>Date Inspection Report Issued To</u>	
		<u>NWB Project Manager</u>	<u>*NWB Management</u>
27-30 Aug 1999	Sample Receipt	30 Aug 1999	31 Aug 1999
08 Sep 1999	Analytical Plan	08 Sep 1999	30 Sep 1999
17 Sep 1999	Sample Analysis	17 Sep 1999	30 Sep 1999
01 Dec 1999	Report Draft/Raw Data	02 Dec 1999	30 Dec 1999
09 Dec 1999	Final Report	09 Dec 1999	30 Dec 1999

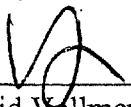
*Reports to NWB Management are issued monthly.

Northwest Bioanalytical

Study No. NWBS99-116
Report No. NWBR99-113

Compliance Statement

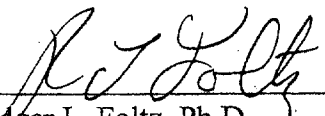
The clinical sample analysis study described within this report is not included within the definition of a GLP regulated nonclinical study set forth in Title 21 of the U.S. Code of Federal Regulations Part 58. However, to the best of our knowledge, this project was conducted in accordance with the guidelines of the U.S. FDA Good Laboratory Practice Regulations for Nonclinical Laboratory Studies (Title 21 CFR Part 58) and according to the methods and procedures described within this report. In addition, the study followed the guidelines of the OECD Principles of Good Laboratory Practice and the Japanese MHW Good Laboratory Practice Standard Ordinance for Nonclinical Laboratory Studies on the Safety of Drugs (Ordinance No. 21, PAB Notification No. 424.) There were no known incidents that affected the quality or integrity of the project or data reported. This report represents an accurate record of the raw data.



David Vollmer, Ph.D.
NWB Project Manager

12/8/99

Date



Rodger L. Foltz, Ph.D.
NWB Laboratory Director

12/9/99

Date

TABLE OF CONTENTS

SIGNATURE PAGE.....	1
QUALITY ASSURANCE STATEMENT	2
COMPLIANCE STATEMENT	3
TABLE OF CONTENTS.....	4
LIST OF TABLES	4
1. INTRODUCTION.....	6
2. METHODOLOGY.....	7
3. SAMPLE ANALYSIS	9
4. RESULTS AND DISCUSSION	10
5. REFERENCES	10

LIST OF TABLES

Table 1. Calibration Curve Summary for PFOS in Human Serum.....	12
Table 2. Calibration Curve Summary for PFOSA in Human Serum.....	12
Table 3. Calibration Curve Summary for PFOSAA in Human Serum	12
Table 4. Calibration Curve Summary for PFHS in Human Serum.....	13
Table 5. Calibration Curve Summary for POAA in Human Serum	13
Table 6. Calibration Curve Summary for M556 in Human Serum.....	13
Table 7. Calibration Curve Summary for M570 in Human Serum.....	14
Table 8. Back-Calculated Concentrations of Calibration Standards for PFOS in Human Serum.....	15
Table 9. Back-Calculated Concentrations of Calibration Standards for PFOSA in Human Serum	15

Table 10. Back-Calculated Concentrations of Calibration Standards for PFOSAA in Human Serum	16
Table 11. Back-Calculated Concentrations of Calibration Standards for PFHS in Human Serum	16
Table 12. Back-Calculated Concentrations of Calibration Standards for POAA in Human Serum	17
Table 13. Back-Calculated Concentrations of Calibration Standards for M556 in Human Serum	17
Table 14. Back-Calculated Concentrations of Calibration Standards for M570 in Human Serum	18
Table 15. Analytical QC Summary for PFOS in Human Serum	19
Table 16. Analytical QC Summary for PFOSA in Human Serum	19
Table 17. Analytical QC Summary for PFOSAA in Human Serum	20
Table 18. Analytical QC Summary for PFHS in Human Serum	20
Table 19. Analytical QC Summary for POAA in Human Serum	21
Table 20. Analytical QC Summary for M556 in Human Serum	21
Table 21. Analytical QC Summary for M570 in Human Serum	22
Table 22. Study Sample Concentrations	23

Quantitative Determination of PFOS, PFOSA, PFOSAA, PFHS, POAA, M556 and M570 in Human Serum by LC/MS/MS

Sample Analysis for Protocol EPI-Swed

1. INTRODUCTION

This report summarizes the analytical results from the quantitation of PFOS, PFOSA, PFOSAA, PFHS, POAA, M556 and M570 in human serum samples for 3M Environmental Technology and Services in support of Protocol EPI-Swed [5.1]. The validated LC/MS/MS method provided a lower limit of quantitation (LLOQ) of 31.4 (PFOS), 1.00 (PFOSA), 1.45 (PFOSAA), 2.61 (PFHS) and 5.27 (POAA) ppb. All analytes exhibited a linear response to approximately 500 ppb with the exception of PFOSAA, which had an upper limit of quantitation (ULOQ) of 270 ppb [5.2]. The non-validated method for the quantitation for M556 and M570 in human serum provided a lower limit of quantitation (LLOQ) of 1.00 and 5.00 ppb, respectively, and exhibited a linear response to 500 ppb for both analytes.

The testing facility was 3M Corporate Occupational Medicine (Bldg. 220-3W-05, St. Paul, MN 55144-1000), and J.H. Mandel, M.D., at the facility served as the Principal Investigator. The following is a list of NWB supervisory personnel involved in the completion of this work: David Vollmer, Ph.D. (NWB Project Manager); Rodger L. Foltz, Ph.D. (NWB Laboratory Director).

NWB SOPs were used in the conduct of this study and were available to study personnel in electronic and/or hard copy formats.

Date Study Initiated: September 7, 1999 Date Analyses Completed: September 20, 1999

The clinical study described in this report is not included within the definition of a GLP regulated nonclinical study. However, Northwest Bioanalytical conducts all studies within the guidelines of the U.S. FDA Good Laboratory Practice Regulations for

Nonclinical Laboratory Studies (Title 21 CFR Part 58), the OECD Principles of Good Laboratory Practice and the Japanese MHW Good Laboratory Practice Standard Ordinance for Nonclinical Laboratory Studies on the Safety of Drugs (Ordinance No. 21, PAB Notification No. 424). Any changes to or deviations from the original protocol (Analytical Plan) were documented through approved protocol amendments or deviation memos and are retained within the raw data.

2. METHODOLOGY

The validated assay used for this study is Meth98082.00 and was reported in Northwest Bioanalytical report NWBR99-005 [5.2].

Samples for this study were received at NWB on the following date:

<i>Receipt Date</i>	<i>Number of Samples Received</i>	<i>Storage Condition (except during analysis)</i>
August 27, 1999	39	- 20°C

Reference Material

<i>Analyte</i>	<i>Lot Number</i>	<i>Purity</i>	<i>Expiration Date</i>	<i>Source</i>	<i>Storage Conditions</i>
*PFOS (FC-95)	193	100%	12/31/2010	3M	Room Temperature (dry)
PFOSA	214	100%	12/31/2010	3M	Room Temperature
*PFOSAA (FC-129)	617	53.8%	12/31/2010	3M	Room Temperature
PFHS	S398-182	100%	12/31/2010	3M	- 20°C
*POAA (FC 143)	245	100%	12/31/2010	3M	Room Temperature (dry)
M556	NB 113047-80	99.89%	12/31/2010	3M	Room Temperature
M570	118506-26	99.75%	12/31/2010	3M	Room Temperature
SDS (sodium dodecylsulfate)	17H0459	99%	1/16/2001	Sigma	Room Temperature

* Analyte names used throughout this report do not include their FC identifier; for example, PFOS is synonymous with PFOS (FC-95)

Nine calibration standards were prepared fresh daily by adding the appropriate spiking standard solution into 0.200 mL of human serum. The final calibration standard concentrations in human serum for each analyte were as follows:

<i>Analyte</i>	<i>Calibration Standard Concentrations (ppb)</i>
PFOS	31.4, 35.4, 40.4, 55.4, 80.4, 130, 280, 430, 530
PFOSA	1.00, 5.00, 10.0, 25.0, 50.0, 100, 250, 400, 500
PFOSAA	1.45, 3.60, 6.29, 14.4, 27.8, 54.7, 135, 216, 270
PFHS	2.61, 6.61, 11.6, 26.6, 51.6, 102, 252, 402, 502
POAA	5.27, 9.27, 14.3, 29.3, 54.3, 104, 254, 404, 504
M556	1.00, 5.00, 10.0, 25.0, 50.0, 100, 250, 400, 500
M570	1.00, 5.00, 10.0, 25.0, 50.0, 100, 250, 400, 500

Analytical QCs were prepared in human serum on September 13, 1999 and frozen in a -20°C freezer. Analytical QCs at the following levels were assayed in duplicate in each analytical run:

<i>Analyte</i>	<i>QC Concentrations (ppb)</i>
PFOS	Low (50.4) and High (380)
PFOSA	Low (20.0) and High (350)
PFOSAA	Low (11.7) and High (189)
PFHS	Low (21.6) and High (352)
POAA	Low (24.3) and High (354)
M556	Low (20.0) and High (350)
M570	Low (20.0) and High (350)

After the addition of HPLC-grade water and sodium dodecyl sulfate (SDS, internal standard) to 0.200 mL of human serum, the serum mixture is made basic with the addition of 0.5 M tetrabutylammonium hydrogen sulfate (TBA, pH 10) and 0.25 M carbonate buffer. This mixture is then extracted with methyl-tert-butyl ether. After sufficient mixing, the sample is centrifuged. The organic layer is transferred via pipette into a clean test tube. The organic layer is then evaporated to dryness and the sample is

reconstituted into 2 mM ammonium acetate water:methanol (50:50 v/v). The extracts are then analyzed by liquid chromatography/tandem mass spectrometry using negative-ion electrospray ionization and multiple reaction monitoring.

3. SAMPLE ANALYSIS

The prepared samples, calibration standards and QCs were injected into the PE Sciex API 3000 LC/MS/MS system in a systematic order.

Chromatographic peak integration was performed using MacQuan software (version 1.6). Quadratic regression analysis (weighted 1/x) for all analytes was performed using the peak area ratio vs. concentration utilizing Watson[®] DMLIMS software (version 5.4.10.2).

3.1. Acceptance Criteria

For an analytical run to be accepted, it must have met the acceptance criteria listed below which are consistent with regulatory and industry recommendations.

Calibration Curve

Each run will include duplicate calibration standards at 7 or more concentrations covering the lower to upper limit of quantitation. At least two-thirds of the calibration standard's back-calculated concentrations must be within $\pm 15\%$ ($\pm 20\%$ for LLOQ) of their individual target concentrations. If a calibration standard is not within the acceptance criteria, it is deactivated. This process starts from the highest calibration standard down to the lowest until all the active standards are within the acceptance criteria.

Lower Limit of Quantitation

The back-calculated concentrations of at least one of the duplicate lowest points in the calibration curve must be within $\pm 20\%$ of the target concentration to qualify as the LLOQ. If this criterion is not met, the next level is subjected to the same test and the LLOQ raised accordingly.

Quality Control Samples

Each analytical run will include low and high QC samples in triplicate. The measured concentrations of at least two-thirds of all analytical QCs must be within $\pm 20\%$ of their target concentrations. If study samples require dilution, a dilution QC is analyzed in duplicate for each dilution level. At least one dilution QC at each level must be within $\pm 20\%$ in order to accept study sample results at that dilution level. The dilution QC acceptance was independent of the undiluted analytical QC acceptance.

4. RESULTS AND DISCUSSION

Each accepted run met the acceptance criteria set for the calibration curve points, for the lower limit of quantitation (LLOQ) and for the analytical quality control (QC) samples.

Run ID	PFOS Result	PFOSA Result	PFOSAA Result	PFHS Result	POAA Result	M556 Result	M570 Result	Comments
1	accepted	accepted	accepted	accepted	accepted	accepted	accepted	PSAE
2	accepted	accepted	accepted	accepted	accepted	accepted	accepted	

PSAE = Pre-study assay evaluation

There were no known circumstances that may have affected the quality or integrity of the data.

In conclusion, based on the quality control and calibration curve data, it is our opinion that the data submitted to 3M Environmental Technology and Services for PFOS, PFOSA, PFOSAA, PFHS, POAA, M556 and M570 are accurate for all of the EPI-Swed study samples. Please note that the results for analytes M556 and M570 should be considered unvalidated data.

5. REFERENCES

- 5.1. 3M Protocol EPI-Swed, "Analysis of Fluorochemicals in Individual Sera from Sweden," August 24, 1999.

- 5.2. D. Vollmer, "Quantitative Determination of PFOS, PFOSA, PFOSAA, N-MeFOSE-OH, N-EtFOSE-OH, POAA and PFHS in Human Serum by LC/MS/MS," NWB Study Number NWBS98-082, NWB Report Number NWBR99-005, May 13, 1999.

The raw data and final report for this study will be stored in the NWB Archives, 1121 East 3900 South, Salt Lake City, UT 84124 per regulations and contract agreement. After submission of the final report to the Sponsor, remaining study samples will be stored under required conditions until confirmation of Sample Disposition/Return Authorization is received from the Sponsor. 3M Environmental Technology and Services and 3M Corporate Occupational Medicine will be notified concerning final disposition of records at completion of contract obligations.

Table 1. Calibration Curve Summary for PFOS in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	-0.000003	0.006330	0.056522	0.9964	31.4	530
Mean		-0.000003	0.006330	0.056522	0.9964		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 2. Calibration Curve Summary for PFOSA in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	-0.000009	0.015340	0.006786	0.9967	1.00	500
Mean		-0.000009	0.015340	0.006786	0.9967		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 3. Calibration Curve Summary for PFOSAA in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	-0.000002	0.001756	0.001162	0.9952	1.45	270
Mean		-0.000002	0.001756	0.001162	0.9952		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 4. Calibration Curve Summary for PFHS in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	-0.000005	0.009627	0.017026	0.9970	2.61	502
Mean		-0.000005	0.009627	0.017026	0.9970		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 5. Calibration Curve Summary for POAA in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	-0.000005	0.011221	0.022280	0.9961	5.27	504
Mean		-0.000005	0.011221	0.022280	0.9961		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 6. Calibration Curve Summary for M556 in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	0.000000	0.000362	0.001145	0.9977	1.00	500
Mean		0.000000	0.000362	0.001145	0.9977		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 7. Calibration Curve Summary for M570 in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	A	B	C	R-Squared	LLOQ	ULOQ
16-Sep-1999	2	-0.000002	0.002831	0.017415	0.9953	5.00	500
Mean		-0.000002	0.002831	0.017415	0.9953		
n		1	1	1	1		

* A, B and C are coefficients used to define the quadratic curve.

Table 8. Back-Calculated Concentrations of Calibration Standards for PFOS in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	31.4	35.4	40.4	55.4	80.4	130	280	430	530
16-Sep-1999	2	*42.2	36.0	42.6	56.5	83.2	119	305	397	534
		28.6	*26.3	40.8	56.1	78.7	131	*340	419	552
Mean		28.6	36.0	41.7	56.3	81.0	125	305	408	543
S.D.		N.C.	N.C.	1.27	0.283	3.18	8.49	N.C.	15.6	12.7
%CV		N.C.	N.C.	3.0	0.5	3.9	6.8	N.C.	3.8	2.3
%Bias		-8.9	1.7	3.2	1.6	0.7	-3.8	8.9	-5.1	2.5
n		1	1	2	2	2	2	1	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 9. Back-Calculated Concentrations of Calibration Standards for PFOSA in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	1.00	5.00	10.0	25.0	50.0	100	250	400	500
16-Sep-1999	2	1.04	*7.31	*11.8	25.3	51.5	87.0	253	354	494
		0.809	4.90	10.8	26.7	51.5	109	*348	419	535
Mean		0.925	4.90	10.8	26.0	51.5	98.0	253	387	515
S.D.		0.163	N.C.	N.C.	0.990	0.00	15.6	N.C.	46.0	29.0
%CV		17.6	N.C.	N.C.	3.8	0.0	15.9	N.C.	11.9	5.6
%Bias		-7.5	-2.0	8.0	4.0	3.0	-2.0	1.2	-3.3	3.0
n		2	1	1	2	2	2	1	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 10. Back-Calculated Concentrations of Calibration Standards for PFOSAA in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	1.45	3.60	6.29	14.4	27.8	54.7	135	216	270
16-Sep-1999	2	1.28	*4.35	6.76	15.1	28.2	48.6	135	185	284
		1.36	3.16	7.06	16.0	28.9	51.5	154	210	285
Mean		1.32	3.16	6.91	15.6	28.6	50.1	145	198	285
S.D.		0.0566	N.C.	0.212	0.636	0.495	2.05	13.4	17.7	0.707
%CV		4.3	N.C.	3.1	4.1	1.7	4.1	9.2	8.9	0.2
%Bias		-9.0	-12.2	9.9	8.3	2.9	-8.4	7.4	-8.3	5.6
n		2	1	2	2	2	2	2	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 11. Back-Calculated Concentrations of Calibration Standards for PFHS in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	2.61	6.61	11.6	26.6	51.6	102	252	402	502
16-Sep-1999	2	2.33	6.97	11.7	27.4	51.3	89.6	257	368	501
		*1.44	*5.13	11.5	28.8	52.3	99.8	281	391	528
Mean		2.33	6.97	11.6	28.1	51.8	94.7	269	380	515
S.D.		N.C.	N.C.	0.141	0.990	0.707	7.21	17.0	16.3	19.1
%CV		N.C.	N.C.	1.2	3.5	1.4	7.6	6.3	4.3	3.7
%Bias		-10.7	5.4	0.0	5.6	0.4	-7.2	6.7	-5.5	2.6
n		1	1	2	2	2	2	2	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 12. Back-Calculated Concentrations of Calibration Standards for POAA in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	5.27	9.27	14.3	29.3	54.3	104	254	404	504
16-Sep-1999	2	5.27	8.96	14.0	29.9	53.9	90.6	260	367	507
		*3.38	*6.93	14.4	32.6	54.8	100	287	399	519
Mean		5.27	8.96	14.2	31.3	54.4	95.3	274	383	513
S.D.		N.C.	N.C.	0.283	1.91	0.636	6.65	19.1	22.6	8.49
%CV		N.C.	N.C.	2.0	6.1	1.2	7.0	7.0	5.9	1.7
%Bias		0.0	-3.3	-0.7	6.8	0.2	-8.4	7.9	-5.2	1.8
n		1	1	2	2	2	2	2	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 13. Back-Calculated Concentrations of Calibration Standards for M556 in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	1.00	5.00	10.0	25.0	50.0	100	250	400	500
16-Sep-1999	2	0.973	5.26	*8.16	25.0	50.0	95.1	244	375	513
		*<LLOQ	*1.84	9.53	26.5	50.5	95.1	281	393	502
Mean		0.973	5.26	9.53	25.8	50.3	95.1	263	384	508
S.D.		N.C.	N.C.	N.C.	1.06	0.354	0.00	26.2	12.7	7.78
%CV		N.C.	N.C.	N.C.	4.1	0.7	0.0	10.0	3.3	1.5
%Bias		-2.7	5.2	-4.7	3.2	0.6	-4.9	5.2	-4.0	1.6
n		1	1	1	2	2	2	2	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 14. Back-Calculated Concentrations of Calibration Standards for M570 in Human Serum

Quadratic weighted 1/x. All concentrations are expressed as ppb.

Run Date	Run Number	1.00	5.00	10.0	25.0	50.0	100	250	400	500
16-Sep-1999	2	*<LLOQ	4.05	10.0	26.1	53.7	89.6	244	350	504
		*<LLOQ	*1.78	10.9	*29.1	54.3	96.6	283	386	558
Mean			4.05	10.5	26.1	54.0	93.1	264	368	531
S.D.			N.C.	0.636	N.C.	0.424	4.95	27.6	25.5	38.2
%CV			N.C.	6.1	N.C.	0.8	5.3	10.5	6.9	7.2
%Bias			-19.0	5.0	4.4	8.0	-6.9	5.6	-8.0	6.2
n			1	2	1	2	2	2	2	2

* Sample deactivated as an outlier ($> \pm 15\%$ theoretical for non-LLOQ; $> \pm 20\%$ theoretical for LLOQ)

N.C. = not calculable

Table 15. Analytical QC Summary for PFOS in Human Serum

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 50.4 ppb	High QC 380 ppb
16-Sep-1999	2	53.2	432
		55.4	416
		55.9	387
Mean		54.8	412
S.D.		1.44	22.8
%CV		2.6	5.5
%Theoretical		108.7	108.4
%Bias		8.7	8.4
n		3	3

Table 16. Analytical QC Summary for PFOSA in Human Serum

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 20.0 ppb	High QC 350 ppb
16-Sep-1999	2	23.6	*448
		19.3	385
		20.1	405
Mean		21.0	413
S.D.		2.29	32.2
%CV		10.9	7.8
%Theoretical		105.0	118.0
%Bias		5.0	18.0
n		3	3

* > $\pm 20\%$ theoretical

Table 17. Analytical QC Summary for PFOSAA in Human Serum

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 11.7 ppb	High QC 189 ppb
16-Sep-1999	2	13.3	218
		*14.4	221
		*14.1	194
Mean		13.9	211
S.D.		0.569	14.8
%CV		4.1	7.0
%Theoretical		118.8	111.6
%Bias		18.8	11.6
n		3	3

* $>\pm 20\%$ theoretical**Table 18. Analytical QC Summary for PFHS in Human Serum**

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 21.6 ppb	High QC 352 ppb
16-Sep-1999	2	21.8	384
		22.7	381
		23.8	330
Mean		22.8	365
S.D.		1.00	30.3
%CV		4.4	8.3
%Theoretical		105.6	103.7
%Bias		5.6	3.7
n		3	3

Table 19. Analytical QC Summary for POAA in Human Serum

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 24.3 ppb	High QC 354 ppb
16-Sep-1999	2	23.1	369
		24.3	381
		25.6	333
Mean		24.3	361
S.D.		1.25	25.0
%CV		5.1	6.9
%Theoretical		100.0	102.0
%Bias		0.0	2.0
n		3	3

Table 20. Analytical QC Summary for M556 in Human Serum

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 20.0 ppb	High QC 350 ppb
16-Sep-1999	2	16.6	374
		19.0	360
		23.8	312
Mean		19.8	349
S.D.		3.67	32.5
%CV		18.5	9.3
%Theoretical		99.0	99.7
%Bias		-1.0	-0.3
n		3	3

Table 21. Analytical QC Summary for M570 in Human Serum

All concentrations are expressed as ppb.

Run Date	Run Number	Low QC 20.0 ppb	High QC 350 ppb
16-Sep-1999	2	22.4	392
		*24.5	382
		22.5	355
Mean		23.1	376
S.D.		1.18	19.1
%CV		5.1	5.1
%Theoretical		115.5	107.4
%Bias		15.5	7.4
n		3	3

* > $\pm 20\%$ theoretical

Table 22. Study Sample Concentrations

All concentrations are expressed as ppb.

Sample ID	PFOS	PFOSA	PFOSAA	PFHS	POAA	M556	M570
9506	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9507	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9508	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9509	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9510	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9511	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9512	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9513	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9514	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9515	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9516	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9517	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9518	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9519	32.1	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9520	31.8	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9521	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9522	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9523	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9524	33.1	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	6.43	<LLOQ(1.00)	<LLOQ(5.00)
9525	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9526	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)

() value in parentheses is the LLOQ

Table 22. Study Sample Concentrations (concentrations)

All concentrations are expressed as ppb.

Sample ID	PFOS	PFOSA	PFOSAA	PFHS	POAA	M556	M570
9527	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9528	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9529	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	6.10	<LLOQ(1.00)	<LLOQ(5.00)
9530	43.3	<LLOQ(1.00)	1.72	<LLOQ(2.61)	7.03	<LLOQ(1.00)	<LLOQ(5.00)
9531	<LLOQ(31.4)	<LLOQ(1.00)	1.57	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9532	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9533	45.9	<LLOQ(1.00)	5.75	<LLOQ(2.61)	5.41	2.92	<LLOQ(5.00)
9534	58.5	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9535	37.6	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	6.15	<LLOQ(1.00)	<LLOQ(5.00)
9536	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9537	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	6.12	<LLOQ(1.00)	<LLOQ(5.00)
9538	85.4	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	12.4	<LLOQ(1.00)	<LLOQ(5.00)
9539	72.9	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	7.42	<LLOQ(1.00)	<LLOQ(5.00)
9540	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9541	31.6	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9542	56.1	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9543	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)
9544	<LLOQ(31.4)	<LLOQ(1.00)	<LLOQ(1.45)	<LLOQ(2.61)	<LLOQ(5.27)	<LLOQ(1.00)	<LLOQ(5.00)

() value in parentheses is the LLOQ