3 4 00687
Site:
Break: 3.4
Other:
3616

FISH SAMPLING ANALYTICAL TECHNIQUES

REMEDIAL INVESTIGATION (RI)/FEASIBILITY STUDY (FS)

McINTOSH PLANT SITE
OLIN CORPORATION
McINTOSH, ALABAMA

for:

Olin Corporation
Charleston, Tennessee

September 1991

Woodward-Clyde Consultants

Consulting Engineers, Geologists, and Environmental Scientists
2822 O'Neal Lane, Baton Rouge, LA 70896

Site:
Break: 3.00668
Other:

Olin CHEMICALS
P.O. BOX 248, LOWER RIVER ROAD, CHARLESTON, TN 37310
PHONE: (615) 336-4000

September 26, 1991

Cheryl Walker Smith
Senior Remedial Project Manager
United States Environmental Protection Agency
345 Courtland Street Northeast
Atlanta, Georgia 30365

Re: Submission of Fish Sampling Analytical Techniques
Olin Chemicals/McIntosh Plant Site
McIntosh, Alabama

Dear Ms. Smith.

Enclosed is the fish sampling analytical techniques, which includes the parameters and methods for analyzing fish from the basin. Fish sampling is scheduled to begin October 7, 1991. Please call me to discuss any issues that you may have regarding this submittal so that we can meet the scheduled start date.

Sincerely,

OLIN CORPORATION

JC Brown jmm

J. C. Brown
Manager, Environmental Technology

jmm
Enclosure

cc: W. A. Beal
D. E. Cooper (2)
W. J. Derocher
M. L. Fries
W. G. McGlasson
J. L. McIntosh
T. B .Odom
R. A. Pettigrew

3 4 00667
Site:
Break: 3-4
Other:
3516

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McINTOSH, ALABAMA

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September 1991

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Appendix B	Mercury Analytical Procedures
Appendix C	Volatile Chlorinated Benzene Analytical Procedures
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1.0 INTRODUCTION

Olin Chemicals (Olin) operates a diaphragm cell chlor-alkali facility in McIntosh, Alabama for the production of chlorine, caustic, sodium hypochlorite and caustic plant (CP) salt. Rocket fuels are also formulated in an onsite blending facility. An Administrative Order by Consent (Consent Order) became effective May 9, 1990 for the performance of Remedial Investigation/Feasibility Study (RI/FS) at the McIntosh Site. Project plans have been prepared and submitted for partial fulfillment of the work items to be performed under the jurisdiction of the Consent Order and include the Work Plan, Health and Safety Plan and Sampling and Analysis Plan.

Two operable units (OU-1 and OU-2) have been designated for the facility. The biological sampling that is described in the project plans will be conducted in OU-2. Operable Unit 2 consists of the basin (65-acre lake), the wetlands within the Olin property boundary and the wastewater ditch leading to the basin. The biological sampling will be performed to obtain the data necessary to evaluate the potential effects of basin contamination on the aquatic biota. The sampling includes a macroinvertebrate survey and fish sampling.

The sample analyses, quality control and reporting for the target parameters in fish are similar to those discussed previously for soil samples in Sampling and Analysis Plan Quality Assurance Project Plan (QAPP) (Volume II of II, Remedial Investigation (RI)/Feasibility Study (FS), McIntosh Plant Site, Olin Corporation, McIntosh, Alabama, May 1991). Details of the fish sampling that is planned for the RI/FS were submitted to EPA in a revised Sampling and Analysis Plan (SAP) on May 5, 1991. As stated in the revised SAP (Section 6.0), the analytical parameters and analytical methods for the fish sampling had not been determined and would be submitted as a separate document.

Since submittal of the revised SAP, preliminary sediment data have been received and these data have been used in selecting the appropriate parameters and analytical techniques for the fish sampling. The fish analytical parameters and techniques are included in this document along with revisions and additions to the May 1991 QAPP that pertain to the fish sampling.

2.0 SAMPLE PARAMETERS

As stated in the revised SAP, the fish tissue will be analyzed for mercury and the organic indicator parameters. The preliminary data from the sediment analyses have been reviewed for the purpose of selecting the appropriate organic parameters.

2.1 Preliminary Sediment Sample Results

Results have been received from 15 sediment samples and one duplicate from the basin and two sediment samples from the former wastewater ditch. These data have not been validated, therefore only a preliminary summary of results reported above the Contract Required Quantitation Limit (CRQL) is presented.

Semivolatile Results

- Phthalates were reported in one sample above the CRQL at a maximum reported concentration of 37 mg/kg. These are not believed to be indicative of the organics in the basin. Phthalate esters are used extensively as plasticizers and commonly show up in environmental analyses resulting from either field or laboratory contaminants. This will be evaluated further during the data validation process.

- Hexachlorobenzene was reported in four samples above the CRQL at a maximum concentration of 40 mg/kg.

- Dichlorobenzenes were reported in one sample above the CRQL at a concentration of 0.47 mg/kg. The dichlorobenzenes were only reported in samples that also showed hexachlorobenzene at significantly higher concentrations. For example, the sample that showed 0.47 mg/kg dichlorobenzenes indicated hexachlorobenzene at a concentration of 20 mg/kg.

- Pentachlorobenzene and pentachloronitrobenzene were reported as tentatively identified compounds.

Volatile Organic Results

- Chlorobenzene was reported in ten samples and the duplicate sample above the CRQL, generally at low concentrations. The maximum concentration reported was 1.0 mg/kg. The sample that showed about 1.0 mg/kg of chlorobenzene also showed hexachlorobenzene at about 40 mg/kg.

- Additional volatiles were reported in the analysis at relatively low concentrations; however, a preliminary review of the results indicates that these are probably laboratory artifacts. A more thorough review of the final data will be conducted during the validation process.

Pesticide/PCB Results

- The most common pesticide/PCB constituents detected were 4,4'-DDE, 4,4'-DDD, and 4,4'-DDT. All three compounds were reported in all 17 samples. The maximum reported for the sum of 4,4'-DDE, 4,4'-DDD, and 4,4'-DDT was about 7.0 mg/kg.

- Endosulfan I and delta-BHC were also reported but were less common and were found at lower concentration (all less than 0.25 mg/kg).

- No other PCB/pesticide compounds were detected above 0.10 mg/kg.

2.2 Selection of Sample Parameters

The criteria for selection of the organic parameters are as follows:

- Constituents most common to the basin based on the preliminary sediment data. These include hexachlorobenzene, 4,4'-DDT, 4,4'- DDD and 4,4'-DDE.

- Constituents most likely to be found and most likely to persist in the basin sediments based on the knowledge of past operations at the facility. These include the chlorinated benzenes that were not reported in sediment results or were reported at low concentrations. Chlorinated benzenes are known to have been present at the Olin facility when wastewater was discharged to the basin. Although they may not be appropriate indicators of sediment contamination, they are selected for the fish analyses due to the potential for bioaccumulation in the fish tissue.

Based on these criteria the analytical parameters selected for the fish analyses are as follows:

Category/Parameter	CLP Target Parameter
Metals	
Mercury	Yes
Volatile Chlorinated Benzenes	
Chlorobenzene (monochlorobenzene)	Yes
Semivolatile Chlorinated Benzenes	
1,2-Dichlorobenzene	Yes
1,3-Dichlorobenzene	Yes
1,4-Dichlorobenzene	Yes
1,2,4-Trichlorobenzene	Yes
Pentachlorobenzene	No
Hexachlorobenzene	Yes
Pentachloronitrobenzene	No

Chlorinated Pesticides	
4-4'-DDT	Yes
4-4'-DDD	Yes
4-4'-DDE	Yes
Lipids	No

3.0 ANALYTICAL METHODS

Tissue sample preparation will be performed by Hazleton Environmental Services (Hazleton) located in Madison, Wisconsin. The preparation techniques are outlined in Appendix A, Preparation of Fish Tissue for Analytical Determinations in the Laboratory, OP-6004-FPREP, Hazleton Environmental Services, Madison, Wisconsin, April 1991.

After completion of the initial fish tissue processing, the following subsamples will be weighed:

Category/Parameter	Sample Weight (grams)	Number of Aliquots
Mercury (Microwave Digestion)	1.0	3
Chlorobenzene	5.0	3
Chlorinated Benzenes, Chlorinated Pesticides, and Lipids	30	3

The sample aliquots will be stored in glass containers with Teflon-lined screw tops. Three subsamples for mercury analysis will be frozen on dry ice and shipped to Olin Chemicals laboratory in Charleston Tennessee for mercury analysis.

The remaining subsamples and bulk sample will be retained by Hazleton. The three subsamples for each category will provide one subsample for the primary analysis and two subsamples held frozen in reserve should reanalysis be required.

The fish tissue analyses will be performed using the following methods.

- **Mercury** (Presented in Appendix B)
  Determination of Total Mercury in Fish, Olin Corporation Product Quality and Environmental Control Department, Charleston, Tennessee, undated.

- **Volatile Chlorinated Benzene** (Presented in Appendix C)
  Method Summary for Chlorobenzene in Fish Tissue by GC/MS, Hazleton Environmental Services, Madison, Wisconsin, September 1991.

- **Semivolatile Chlorinated Benzenes, Chlorinated Pesticides and Lipids** (Presented in Appendix D)
  Semivolatile Analysis of Fish Samples by Gas Chromatography/Mass Spectroscopy (GC/MS), MP-HZB-MA, Hazleton Environmental Services, Madison, Wisconsin, undated.

4.0 DETECTION LIMITS

The method detection limits for the fish analytical parameters are as follows:

Category/Parameter	Method Detection Limit mg/kg
Metals	
Mercury	0.10
Volatile Chlorinated Benzenes	
Chlorobenzene (monochlorobenzene)	0.005
Semivolatile Chlorinated Benzenes	
1,2-Dichlorobenzene	0.660
1,3-Dichlorobenzene	0.660
1,4-Dichlorobenzene	0.660

1,2,4-Trichlorobenzene	0.660
Pentachlorobenzene	0.660
Hexachlorobenzene	0.660
Pentachloronitrobenzene	0.660
Chlorinated Pesticides	
4,4'-DDT	0.660
4,4'-DDD	0.660
4,4'-DDE	0.660
Lipids	Not Applicable

Elevated detection limits are anticipated only for those samples exhibiting high analyte concentration levels and/or interferences.

5.0 SAMPLE PRESERVATION AND HOLDING TIMES

As stated in the revised SAP, the fish samples will be placed in glass jars with teflon-lined screw tops and will be shipped from the field to the laboratory protected from light and at wet ice temperature. The samples will be stored, protected from light and frozen at approximately -20°C from the time of receipt in the laboratory until sample preparation and analysis.

There are no established holding times for the target analytes in fish. The analysis for chlorobenzene will be performed within 7 days of sample preparation.

6.0 QUALITY CONTROL

The quality control analyses will include method blanks, duplicates and matrix spikes. The quality control analyses for the chlorinated benzenes will also include internal standards and surrogates.

A method blank will be processed each time 20 or fewer samples are prepared or a new source of reagent is utilized.

A laboratory duplicate and matrix spike will be analyzed for each group of 10 or fewer mercury field samples. The control limit for the matrix spike recovery is 70 to 130 percent.

A laboratory matrix spike and matrix spike duplicate will be analyzed for each group of 20 field samples for chlorobenzene, the semivolatile chlorinated benzenes and chlorinated pesticides. The control limits for the chlorobenzene will correspond to those listed for chlorobenzene in soil in the May 1991 QAPP. The control limits for the chlorinated benzenes and chlorinated pesticides will be as follows:

Parameter	Percent Recovery	Relative Percent Difference
1,2-Dichlorobenzene	10-90[1]	49[1]
1,3-Dichlorobenzene	10-90[1]	49[1]
1,4-Dichlorobenzene	10-90	49
1,2,4-Trichlorobenzene	10-120	33
Pentachlorobenzene	10-120[1]	33[1]
Hexachlorobenzene	10-120[1]	33[1]
Pentachloronitrobenzene	10-120[1]	33[1]
4,4'-DDT	10-120[1]	49[1]
4,4'-DDD	10-120[1]	49[1]
4,4'-DDE	10-120[1]	49[1]

[1] Estimated.

7.0 REPORTING

The final analytical reports will include the following:

Case Narrative
Chain-of-Custody
Quality Control Summary
Sample Data
Calibration
Raw Quality Control Data
Sample Preparation Logs
Instrument Run Logs

8.0 DATA VALIDATION

Data validation will be performed in a manner similar to that used for the Contract Laboratory (CLP) data as described in Sample Analysis Plan Quality Assurance Project Plan (QAPP) (Volume II of II, Remedial Investigation (RI/Feasibility Study (FS), McIntosh Plant Site, Olin Corporation, McIntosh, Alabama, May 1991).

Appendix A

APPENDIX A

FISH TISSUE SAMPLE PREPARATION

TECHNICAL OPERATING PROCEDURE

PROCEDURE TITLE: Preparation of Fish Tissue for Analytical Determinations in the Laboratory

AREA OF APPLICABILITY: Hazleton Wisconsin, Inc.
Environmental Chemistry

SCOPE:

This document outlines the preparation of Fish samples prior to laboratory analysis. These preparation steps are compatible with the analysis of volatile organics, semivolatile organics, Pesticides/PCBs and the determination of heavy metals. These procedures may also be used in the preparation of other biological organisms for the same parameters.

EQUIPMENT AND MATERIALS:

- Teflon-coated or porcelain spatula
- Pyrex glass tray, 8x12x2 inch.
- Knife, heavy blade (or meat cleaver)
- Mallet, plastic faces, 2 to 3 lbs.
- Hobart Stainless Steel meat grinder
- Blender, stainless steel blade (glass or stainless steel blender container only, No plastic containers)
- 40 ml glass vials with teflon septum caps
- Wide-mouth 8-16 oz glass jars with teflon lined caps
- Dry ice or liquid Nitrogen
- Methanol
- Hexane, Pesticide grade

PROCEDURE:

Note: Preparation steps must be done in a clean environment (i.e. not in an extraction lab) when the determination of volatile organics is to be determined. All efforts should be made to minimize sources of contamination.

1.0 All equipment to be used in the preparation of tissue samples should be soap and water washed and then sequentially rinsed with hot water, milli-Q water, methanol, and hexane. This procedure should be repeated between samples before re-using the equipment.

2.0 Fish samples may be prepared with the following specifications:

- Whole Fish
- Fillet of Fish
- Skin On
- Skin Off

The type of preparation usually takes into account, the size of the fish, the species being investigated and the class of the compound or compounds to be determined. Be sure to clarify the specific requirements of the project sponsor before proceeding in the preparation of the fish.

3.0 To prepare the fish sample for analytical pretreatment, unwrap and weigh each fish. (Small fish, such as minnows are usually collected as composites and will represent a single composite sample). A total weight of 250 grams is the perferred mass required when a complete screening of the fish is to be performed (Volatile organics, Semivolatile organics, Pesticides, Metals).

4.0 Chop larger fish into 2 to 3-inch cubes, using either a sharp knife and mallet or a butcher saw.

5.0 Grind the fish cubes in a large commercial meat grinder that has been precooled by grinding dry ice or by rinsing with liquid Nitrogen. (Note: When dealing with small quantities of tissue such as minnows, use a blender instead of the meat grinder, to minimze the loss of tissue in the grinding process itself).

6.0 Thouroughly mix the ground material, using a porcelain or teflon coated spatula. Regrind and mix material two additional times. Clean out any material remaining in the grinder; add this to the sample and mix well.

7.0 Weigh three 5.0 oz portions of the sample into 40 ml glass vials recording the tare weight of the vial and the final weight of the vial plus sample. Cap the vial with a teflon septum screw cap and store in a freezer until ready for volatile analysis.

8.0 Transfer the remaining fish sample to a glass container and store in a freezer for later subsampling and analysis.

APPROVED BY: _____ DATE _____

David C. Hills
Manager
Environmental Chemistry

REVIEWED BY: _____ DATE _____

Deborah L. Keller
Manager
Quality Assurance Unit

Appendix B

APPENDIX B

MERCURY ANALYTICAL PROCEDURES

OLIN CORPORATION
PRODUCT QUALITY AND ENVIRONMENTAL CONTROL DEPARTMENT
CHARLESTON, TENNESSEE

DETERMINATION OF TOTAL MERCURY IN FISH

1.0 THEORY

1.1 This is a cold-vapor atomic absorption procedure at 253.7-nm. wavelength for the determination of total mercury in fish. After digestion, the mercury is reduced to the elemental state and aerated from solution in an open system. The mercury is passed through a cell positioned in the light path of an atomic absorption spectrometer. The absorbance (peak height) is measured as a function of mercury concentration.

Also, the method can be utilized for total mercury analyses of other biota.

2.0 SAFETY

2.1 Familiarize yourself with all equipment and chemicals used in this procedure according to the manufacturer's instructions.
Maintain an awareness of the danger associated with the various types of reagent chemicals used in this procedure.

2.2 All spills must be cleaned immediately and in the case of skin contact, wash with large volumes of cold water.

2.3 Because mercury vapor is toxic, precaution must be taken to avoid its inhalation. Therefore, the mercury vapor discharge from the cell must be vented into an exhaust hood or be passed through some type of absorbing medium.

2.4 Exhaust hood used for the digestion procedure must be approved for perchloric acid fumes.

3.0 APPARATUS

3.1 Atomic absorption spectrophotometer or equivalent: any atomic absorption spectrophotometer with an open sample presentation area in which to mount the absorption cell is suitable. Commercial instruments

are available using the cold-vapor technique and may be substituted for the atomic absorption spectrophotometer.

3.2 Exhaust hood: An approved perchloric acid hood.

3.3 Hot Plate: Variable thermostatic controller with a specific power of at least 1.7 watts/cm^2.

3.4 Flowmeter: Capable of measuring an air flow of one-liter/min.

3.5 Drying tube: 6-inch x 3/4-inches in diameter tube containing 10-20 grams of magnesium perchlorate.

3.6 Extension column: 7 1/4 inches long with 19/38 ground glass fitting.

3.7 Volumetric flask: 100 ml with 19/38 ground glass fitting.

3.8 BOD flask: 250 ml or equivalent with proper ground glass fitting.

3.9 Pipettes: Various sizes of glass pipettes

3.10 Recorder: Any multirange variable-speed recorder that is compatible with the atomic absorption UV detector system.

3.11 Cold-vapor Generator: See diagram 3.11

4.0 <u>REAGENTS</u>

4.1 Aqua Regia: Prepare daily three volumes of concentrated hydrochloric acid and one volume of concentrated nitric acid.

4.2 Nitric Acid, HNO_3: Concentrated, Reagent Grade

4.3 Perchloric Acid, $HClO_4$: 98 Percent, Reagent Grade

4.4 Hydrochloric Acid, HCL: Concentrated, Reagent Grade

4.5 D.I. Water, ASTM Type II

4.6 Potassium Dichromate, $K_2Cr_2O_7$: .02 Percent W/V, dissolve 0.2 grams of $K_2Cr_2O_7$ in D.I. Water and dilute to 1000 mL volume with D. I. water.

4.7 Stannous Chloride, $SnCl_2$: 10 Percent W/V; dissolve 119 grams of reagent grade $SnCl_2$ $2H_2O$ in 40 mL of concentrated hydrochloric acid and dilute to a liter volume with D.I. water.

4.8 Potassium Permanganate, $KmNO_4$: 5% W/V; dissolve 50 grams of $KmNO_4$ in 1,000 mL D. I. Water.

4.9 Magnesium perchlorate, Mg $(ClO_4)_2$; Anhydrous reagent grade crystals.

4.10 Mercury Standard, Hg: 1,000 μg/mL, dissolve 1.354 grams of $HgCl_2$ in 750 mL of D. I. water. Add 100 mL of concentrated nitric acid and dilute to a one-liter volume with D. I. Water. Commercial mercury standards are available and may be substituted for the above mercury standard.

4.11 Hydroxylamine hydrochloride, H_2NOH HCl: 4% W/V, dissolve 20 grams of reagent grade H_2NOH HCl in 60-70 mL D.I. Water. Adjust the pH of the solution to 2.8 + .2 with ammonium hydroxide, NH_4OH. Add 10 mL of a 4% sodium diethyldithiocarbamate solution to complex any metallic impurities. After five minutes, extract the metallic impurities with 20 mL of chloroform in a separatory funnel. Repeat the chloroform extraction twice discarding the chloroform layer after each extraction. Adjust the pH of the aqueous layer to 1.2 with hydrochloric acid and dilute to 500 mL volume with D.I. Water.

4.12 Absorbing Medium for mercury removal:

A. 0.25% (W/V) - iodine in a 3% (W/V) potassium iodide solution, or

B. equal volumes of 0.1 M $KmNO_4$ solution and 10% (W/V) H_2SO_4 Solution.

5.0 PROCEDURE

5.1 Standard Procedure

Prepare daily a 10μg/mL mercury solution by diluting a 10 mL aliquot of the 1,000 μg/mL mercury standard solution and 15 mL of concentrated nitric acid to a liter volume with D.I. Water.

Prepare a 0.1 μg/mL mercury solution by diluting a 1 mL aliquot of the 10μg/mL mercury solution and 15 mL of concentrated nitric acid to a volume of 100 mL with D.I. Water. Transfer 0, 0.5, 1, 3, and 5 mL aliquots of the 0.1 μg/mL mercury solution into a series of BOD flasks. Add 5 mL of freshly prepared aqua regia, 50 mL D.I. Water, 15 mL of $KmNO_4$ solution to each flask and heat thirty minutes in a water bath at 95 degrees Celsius. Allow the solutions to cool and add 6 mL of hydroxylamine hydrochloride solution to reduce the excess potassium permanganate to each flask. Add enough D.I. water to each flask to yield a total volume of 125 mL and analyze each aliquot by cold-vapor procedure.

5.2 Sample Preparation

Samples are individually filleted. Filet only is retained and kept frozen until ready for analysis. Allow filet to reach room temperature, place in a 250 ml beaker and homogenize thoroughly using Tissuemizer. Clean all associated equipment with 10 percent nitric acid to eliminate elevated cross-contamination after each sample is tissuemized.

5.3 Digestion Procedure

Preheat three hot plates in an approved perchloric acid exhaust hood to surface temperatures of 230, 265, and 340 $\pm$5 degrees Celsius respectively.

Weigh approximately three grams of filet to the nearest .01 grams into a 100 mL volumetric flask. Add one mL of the $K_2Cr_2O_7$ Solution, 15 mL HNO_3, and 15 mL $HClO_4$ to each aliquot flask, mixing gently between each addition. Attach an extension column and place the apparatus on the first hot plate (See Diagram 5.2 attached) with a surface temperature of 230 degrees centigrade. Allow the apparatus to remain for 20-30 minutes or until all Nitrous Oxide fumes have visually ceased to evolve.

Place the apparatus on the second hot plate (surface temperature of 265 degrees Celsius) and allow it to remain there until a color change from yellow to green occurs.

Place the apparatus on the third hot plate (surface temperature at 340 degrees Celsius) and allow it to remain there until a color change from green to orange occurs.

Remove the apparatus from the hot plate and let cool to room temperature.

Remove the extension column and wash with D.I. water, collecting the washings in the 100 mL volumetric flask.

Bring to a volume of 100 mL with D.I. Water.

Pipette an aliquot containing an estimated .05 - .5 μg of mercury into a BOD flask. Add enough D.I. Water to make a total volume of 125 mL. The aliquot is now analyzed by cold-vapor atomic absorption spectrophotometer.

5.4 Auto-Digestion Procedure

The Charleston laboratory utilizes a Prolabo "Microdigest 300" microwave system which consists of an automated, computer controlled, 16-sample turntable capable of controlling various digestion parameters within very close tolerance.

Single laboratory analytical comparison studies between these two digestion procedures on forty-one samples with a range of 0.25 - 2.0 mg/kg showed a -0.025 mg/kg bias and 0.99 correlation coefficient. Also, a certified SRM (DORM-1) sample was analyzed which showed a 98.2 percent accuracy, -0.015 mg/kg bias and 6.4 percent RSD.

Weigh approximately one gram of filet to the nearest 0.01 grams into an auto-digestion flask, connect condenser, and place the flask into the sample turntable.

A duel treatment process (digital storage: SRM7B) utilizes a 3 ml HNO_3, 5 ml H_2SO_4, 5 percent power rate for seven minutes and a 1 ml $HClO_4$ and 5 percent power rate for three minutes.

Refer to the Operation Manual for complete operation of the Prolabo "Microdigest 300."

After digestion is completed, dilute to a 50 ml volume with DI water.

Pipette an aliquot containing an estimated .05 - .5 µg of mercury into a BOD flask. Add enough D.I. Water to make a total volume of 125 mL. The aliquot is now analyzed by cold-vapor atomic absorption spectrophotometer.

5.5 Cold-Vapor Procedure

Add 5 mL of stannous chloride solution; immediately attach the BOD bottle to the aeration apparatus. The sample is allowed to stand quietly without manual agitation. The nitrogen purge which has previously been adjusted to a rate of one liter/minute is allowed to purge the sample continuously. The absorbance will increase and reach a maximum within 30 seconds.
As soon as the recorder base line or levels pen levels off (approximately one minute), disconnect BOD bottle and continue to purge the cell with Nitrogen. Record the maximum peak height and calculate the concentration of mercury from the predetermined five-point calibration curve.

6.0 CALCULATIONS

6.1 Measure the peak height of the unknown from the chart and read the mercury value from the standard curve.

6.2 Calculate the mercury concentration in the sample by the formula:

$$\mu g \text{ Hg/gram} = \frac{\mu g \text{ Hg in aliquot}}{\text{wt. of aliquot in gms}}$$

6.3 Report mercury concentrations as follows:

Below 0.1 µg/gm, <0.1 µg; between 0.1 and 1 µg/gm, to nearest 0.01 µg; between 1 and 10 µg/gm, to nearest 0.1 µg; above 10 µg/gm, to nearest µg.

7.0 QUALITY ASSURANCE

7.1 All quality assurance data should be maintained, available for easy reference or inspection, and reported with the results.

7.2 A reagent blank must be analyzed with each batch or every twenty samples whichever is greater.

7.3 A sample duplicate must be analyzed with each batch or grouping of ten (10) samples or less.

7.4 A sample spike must be analyzed with each batch or with each grouping of ten (10) samples or less. Percent recovery for the spikes should be within the 70 - 130 percent range.

7.5 A check standard should be analyzed at the beginning of a batch of samples and after each set of ten sample runs. The percent delta or change in concentration should be less than 15 percent between each check standard.

Written by: H B Cochran
H. B. Cochran

Approved by: J P Newman
J. P. Newman

4.HBC/cb

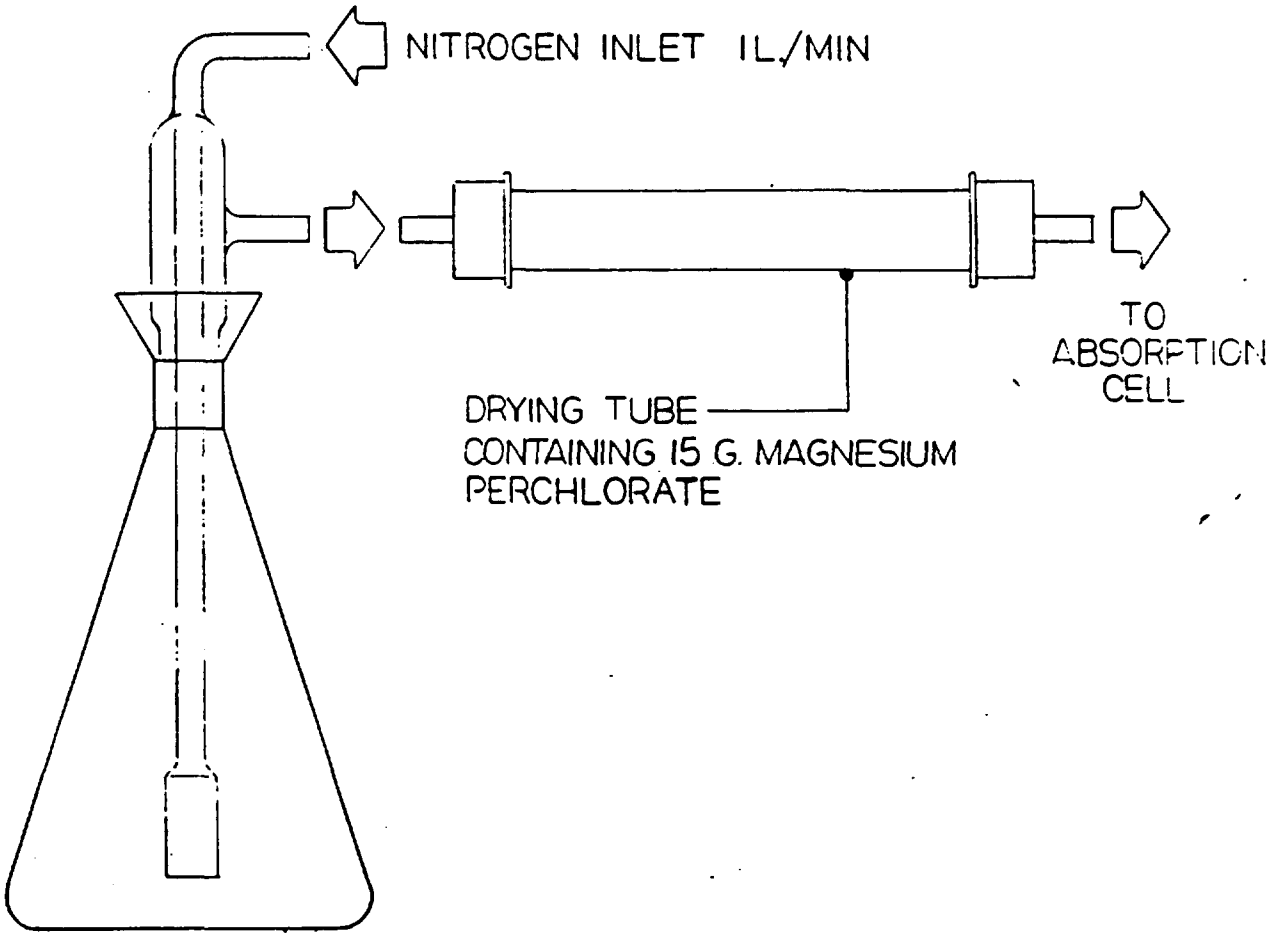
NITROGEN INLET 1L/MIN
TO ABSORPTION CELL
DRYING TUBE CONTAINING 15 G. MAGNESIUM PERCHLORATE

Diagram 3.11

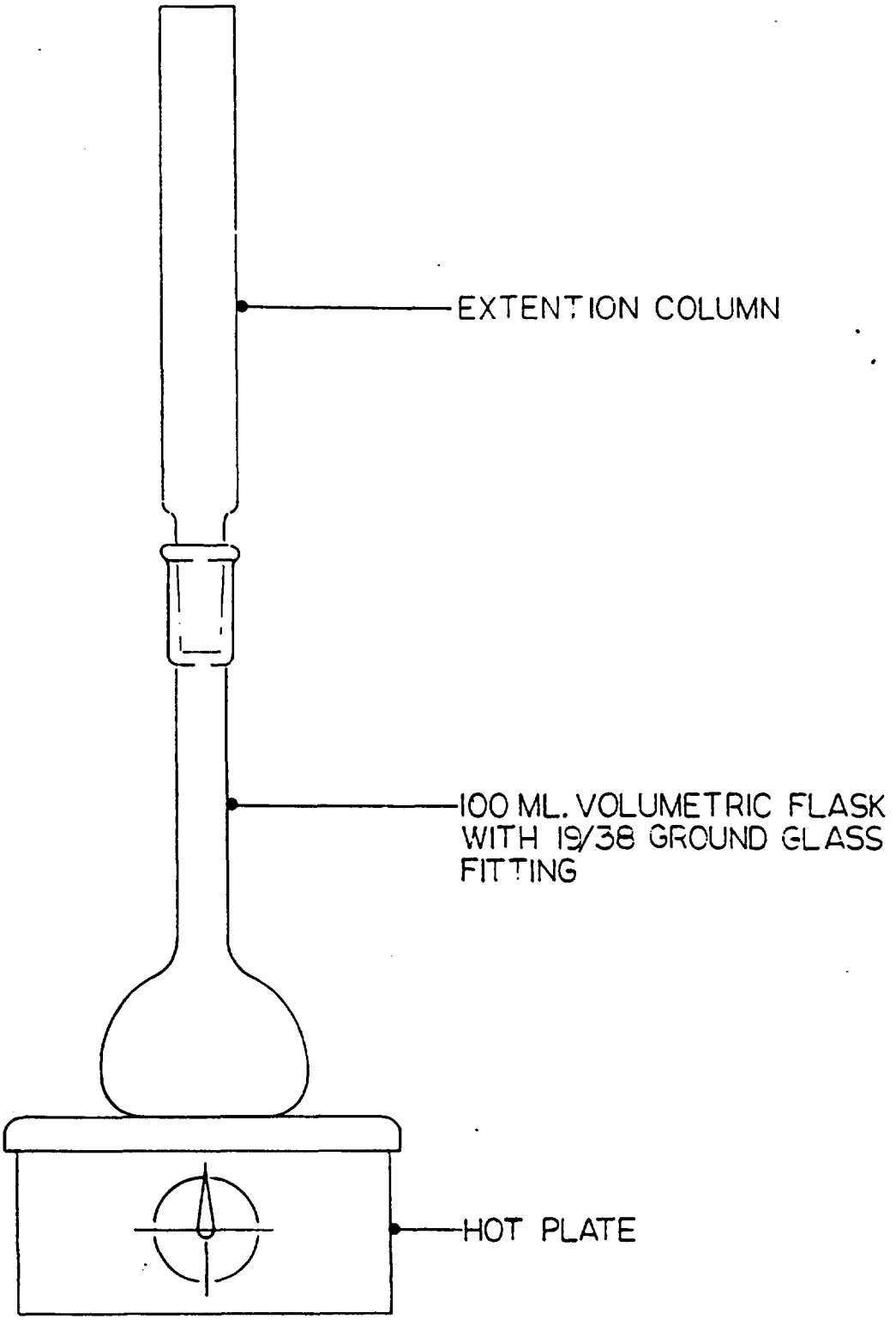
EXTENTION COLUMN
100 ML. VOLUMETRIC FLASK
WITH 19/38 GROUND GLASS
FITTING
HOT PLATE

Diagram 5.2

Appendix C

APPENDIX C

VOLATILE CHLORINATED BENZENE ANALYTICAL PROCEDURES

HAZLETON
ENVIRONMENTAL SERVICES
525 SCIENCE DRIVE
MADISON, WISCONSIN 53711

3 4 00713

a CORNING Laboratory Services Company

September 19, 1991

Method Summary for Chlorobenzene in Fish Tissue by GC/MS

The analysis of chlorobenzene in fish tissue is performed in accordance with the US EPA 2/88 CLP protocol for soil analysis with modifications as follows:

Sample Preparation: All samples will be stored in a walk in freezer until sample preparation is performed. The individual fish samples will be homogenized in order to obtain a representative sample. The homogenizing will be performed with dry ice / liquid nitrogen to reduce volatilization that could occur during sample preparation.

Sample Analysis: Each sample will have 10uL of food grade anti-foam agent added to the sample immediately prior to analysis. This will not introduce any chlorobenzene to the analysis.

Only one internal standard, d5-Chlorobenzene, will be used for quantitation purposes. Only one surrogate standard will be utilized, bromofluorobenzene, with the recovery range equal to the recovery range of soil.

The samples will have matrix spike / matrix spike duplicate performed using only chlorobenzene as the spiking compound. The QC criteria will remain consistent with EPA CLP soil limits.

Detection Limit: The detection limit for chlorobenzene in fish is 5 ug/Kg.

REFERENCE:

1. Environmental Protection Agency Contract Laboratory Program Statement of Work for "Organic Analysis Multi-Media Multi-Concentration, " (February 1988). Exhibits B, D, E, Volatile Sections.

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Appendix D

APPENDIX D

SEMIVOLATILE CHLORINATED BENZENE, CHLORINATED PESTICIDES AND LIPIDS ANALYTICAL PROCEDURES

ASSAY TITLE: Semivolatile Analysis of Fish Samples by Gas Chromatography/Mass Spectroscopy (GC/MS)

AREA OF APPLICABILITY: Hazleton Wisconsin, Inc. Environmental Analysis

SCOPE:

This method is applicable to the determination of residues of the U.S. Environmental Protection Agency (EPA) - defined priority pollutants and target compound list (TCL) semivolatile organic compounds in fish and other aquatic biota. Analysis is conducted using GC-MS. Applicable analytes and their target detection limits are listed in Attachment 1.

PRINCIPLE:

Aliquots of homogenized sample are spiked with a surrogate standard mixture, dried by mixing with anhydrous sodium sulfate, and extracted with methylene chloride in soxhlet extractors. The methylene chloride extracts are purified by gel permeation chromatography (GPC), followed by florisil chromatography. The extracts are then concentrated, and an internal standard mixture is added as a quantification aid for subsequent analysis by GC-MS. Analytes are quantified versus calibration factors obtained from concurrently analyzed standards. GC-MS calibration is done according to guidelines given in the protocols of the EPA Contract Laboratory Program.

SENSITIVITY:

The sensitivity of this method depends on the level of interference within a given matrix.

Target quantification limits are listed in Attachment 1.

PRECISION AND ACCURACY:

The method is capable of providing precise and accurate determinations of analytes as summarized in Attachment 2 for spiked control fish. The data in Attachment 2 were acquired along with analysis of field samples between November 1987 and November 1988.

REFERENCES:

Federal Register, 49(209): 43385-43406, Environmental Protection Agency (EPA) Method 625 (October 16, 1984).

EPA Contract Laboratory Program, Statement of Work for "Organic Analysis Multi-Media Multi-Concentration" (October 1986). Revisions: January 1987; February 1987; July 1987; August 1987. Exhibits: B, D, E.

Methods 3540, 3640, and 8270, "Test Methods for Evaluating Solid Waste," EPA Publication SW-846, Third Edition, Washington, DC, Rev. 0 (September 1986).

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SAFETY PRECAUTIONS:

- The toxicity and carcinogenicity of the chemicals used in this method have not been precisely defined; each chemical should be treated as a potential health hazard and exposure to these chemicals should be minimized. Each analyst is responsible for maintaining awareness of OSHA regulations regarding safe handling of chemicals used in this method. Additional references to laboratory safety are available for the information of the analyst.

- The following parameters covered by this method have been tentatively classified as known or suspected human or mammalian carcinogens: benzo(a)anthracene, benzidine, 3,3'-dichlorobenzidine, benzo(a)pyrene, dibenzo(a,h)anthracene, and N-nitrosodimethylamine. Primary standards of these toxic compounds should be prepared in a hood. A NIOSH/MESA approved toxic gas respirator should be worn when the analyst handles high concentrations of these toxic compounds.

- Observe all standard laboratory safety procedures as outlined in the Hazleton Wisconsin safety training manual.

INTERFERENCES:

Method interferences may be caused by contaminants in solvents, reagents, glassware, and other sample-processing hardware that lead to discrete artifacts or elevated baselines in the total ion current profiles (TICPs). All of these materials must be routinely demonstrated to be free from interferences under the conditions of the analysis by running laboratory reagent blanks. Matrix interferences may be caused by excess lipids in the sample extract or by contaminants that are extracted from the sample. The extent to which the matrix causes interferences will vary considerably from source to source. Cleanup procedures provided in this method may be used to minimize such interferences.

QUALITY ASSURANCE:

This section outlines the minimum quality control operations necessary to satisfy the analytical requirements associated with the determination of semivolatile organic TCL compounds in water.

1. Tuning and GC/MS mass calibration

   It is necessary to establish that a given GC/MS meets the standard mass spectral abundance criteria before initiating any ongoing data collection. This is accomplished through the analysis of decafluorotriphenylphosphine (DFTPP).

1.1 Each GC/MS system used for analysis of semivolatile compounds must be hardware-tuned to meet the abundance criteria listed in Attachment 3 for a 50-ng injection of DFTPP. DFTPP may be analyzed separately or as part of the calibration standard. The criteria must be demonstrated daily or for each 12-hour period, whichever is more frequent, before samples can be analyzed. DFTPP must be injected to meet this criterion.

1.2 Whenever corrective action is taken that may change or affect the tuning criteria for DFTPP (e.g., ion source cleaning or repair, etc.), the tuning must be verified regardless of the 12-hour tuning requirements.

2. Calibration of the GC/MS system

Before the analysis of samples and required blanks and after meeting the tuning criteria, the GC/MS system must be initially calibrated at a minimum of three concentrations to determine the linearity of response using TCL compound standards. Once the system has been calibrated, the calibration must be verified each 12-hour time period for each GC/MS system.

2.1 Prepare calibration standards containing all semivolatile target compounds (Attachment 10) at concentrations of 20, 50, and 100 µg/mL. Initial calibration is then performed by injecting 1 µL of each standard concentration, producing on-column injections of 20, 50, and 100 total ng.

2.2 Analyze each calibration standard and tabulate the area of the primary characteristic ion (Attachments 4 and 5) against concentration for each compound, including all required surrogate compounds. The relative retention times of each compound in each calibration run should agree within 0.06 relative retention time limits. Late-eluting compounds will usually have much better agreement.

2.2.1 Using Attachment 6, calculate the relative response factor (RRF) for each compound at each concentration level using the following Equation.

$$RRF = \frac{A_x}{A_{is}} \times \frac{C_{is}}{C_x}$$

Where:

A_x = Area of the characteristic ion for the compound to be measured.

A_{is} = Area of the characteristic ion for the specific internal standards from Attachment 4 or 5.

C_{is} = Concentration of the internal standard (ng/µL).

C_x = Concentration of the compound to be measured (ng/µL).

2.2.2 Using the RRFs from the initial calibration, calculate the percent of relative standard deviation (%RSD) for compounds labeled as 2 calibration check compounds (CCCs) and shown in Attachment 6 using the following equation.

$$\% RSD = \frac{SD}{\bar{X}} \times 100$$

Where:

RSD = relative standard deviation

SD = standard deviation of initial response factors (per compound)

Where: $$SD = \sqrt{\sum_{i=1}^{N} \frac{(x_i - \bar{X})^2}{N-1}}$$

$\bar{X}$ = Mean of initial relative response factors (per compound)

2.2.3 The %RSD for each CCC (Attachment 7) must be <u>less</u> than or equal to 30.0%. These criteria must be met for the initial calibration to be valid.

2.3 A system performance check must be done to ensure that minimum average RRF are met before the calibration curve is used.

2.3.1 For semivolatiles, the system performance check compounds (SPCCs) are N-Nitroso-Di-n-Propylamine, Hexachlorocyclopentadiene, 2,4-Dinitrophenol, and 4-Nitrophenol. The minimum acceptable average RRF for these compounds is 0.050. SPCCs typically have very low RRFs (0.1 to 0.2) and tend to decrease in response as the chromatographic system or the standard material begins to deteriorate. These compounds are usually the first to show poor performance. Therefore, they must meet the minimum requirement when the system is calibrated.

2.3.2 The initial calibration is valid only after both the %RSD for CCCs and the minimum RRF for SPCCs have been met. Only after both these criteria are met can sample analysis begin.

2.4 Continuing calibration

Continuing calibration is performed by injecting 1 µL of a 50 µg/mL standard containing all semivolatile TCL compounds, including all required surrogates, and is performed every 12 hours when samples are analyzed. Compare the RRF data from the standard every 12 hours with the average RRF from the initial calibration for a specific instrument. A system performance check must be made every 12 hours. If the SPCC criteria are met, a comparison of relative response factors is made for all compounds. This is the same check that is applied during the initial calibration. If the minimum RRFs are not met, the system must be evaluated and corrective action must be taken before sample analysis begins.

2.4.1 Some possible problems are standard mixture degradation, injection port inlet contamination, contamination at the front end of the analytical column, and active sites in the column or chromatography system. This check must be met before analysis begins. The minimum RRF for semivolatile SPCC is 0.050.

2.4.2 Calibration check compounds (CCCs)

After the system performance check is met, the CCCs listed in Attachment 7 are used to check the validity of the initial calibration. Calculate the percent difference using the following Equation.

$$\% \text{ Difference} = \frac{RRF_I - RRF_c}{RRF_I}$$

Where:

RRF_I = Average response factor from initial calibration

RRF_c = Response factor from current verification check standard.

2.4.3 If the percent difference for any compound is greater than 20%, this should be considered a warning limit. If the percent difference for each CCC is less than or equal to 30%, the initial calibration is assumed to be valid. If the criteria are not met (>30% difference) for any CCC, corrective action <u>must</u> be taken.

2.5 Documentation

Calculate and report the RRF and percent difference (%D) for each compound. Ensure that the minimum RRF for semivolatile SPCCs is <u>0.050</u>. The %D for each CCC compound must be less than or equal to 30.0%.

3. Internal standard evaluation

Internal standard responses and retention times in all samples must be evaluated during or immediately after data acquisition. If the retention time for any internal standard changes by more than 30 seconds from the latest daily (12-hour) calibration standard, the chromatographic system must be inspected for malfunctions and corrections must be made as required. The extraction ion current profile (EICP) area of the internal standards must be monitored and evaluated for each sample, blank, matrix spike, and matrix spike duplicate. If the EICP area for any internal standard changes by more than a factor of two (-50% to +100%), the mass spectrometric system must be inspected for malfunction and corrections must be made as appropriate. When corrections are made, the samples that were analyzed while the system was malfunctioning must be reanalyzed.

4. Method blank analysis

A method blank is a 30 g sample of sodium sulfate that is carried through the entire analytical scheme (extraction, concentration, and analysis). This information should be well documented and kept on file.

A method blank is analyzed with every 20 samples processed or whenever samples are extracted, whichever is most frequent. It is the analyst's responsibility to ensure that method interferences caused by contaminants in solvents, reagents, and glassware are minimized. An acceptable laboratory blank should contain less than or equal to the reported method detection limit of any single target compound, or in the case of the phthalate esters, less than or equal to five times (5x) the respective detection limit.

If a laboratory method blank exceeds the above criteria, the analyst must consider the analytical system to be out of control. The source of the contamination must be investigated, and appropriate corrective action must be taken.

5. Surrogate spike (SS) analysis

Surrogate standard recoveries are determined for all samples and blanks. All samples and blanks are fortified with surrogate spiking compounds before extraction in order to monitor sample preparation and analysis.

5.1 Each sample, matrix spike, matrix spike duplicate, and blank are spiked with surrogate compounds before extraction. The surrogate spiking compounds shown in Attachment 8 are used to fortify each sample, matrix spike, matrix spike duplicate, and blank with the proper concentrations.

5.2 Surrogate spike recovery must be evaluated by determining whether the concentration (measured as percent recovery) falls inside the recovery limits listed in Attachment 9.

5.3 If the recovery of any surrogate compound in <u>either</u> base neutral or acid fraction is below 10%, or if the recoveries of two surrogate compounds in <u>either</u> base neutral or acid fractions are outside surrogate spike recovery limits, the analyst will document (i.e., record and discuss the problem and corrective action taken in the case narrative) and deviations from acceptable quality control limits and take the following actions:

5.3.1 Check calculations to ensure that there are no errors; check internal standard and surrogate spiking solutions for degradation, contamination, etc.; and check instrument performance.

5.3.2 If Step 5.3.1 fails to reveal a problem, then reanalyze the extract. If reanalysis of the extract solves the problem, then the problem was within the analyst's control. Therefore, only submit data from the analysis with surrogate spike recoveries within the control windows. This will be considered the initial analysis and will be reported as such on all data deliverables.

5.3.3 If Step 5.3.2 fails to solve the problem, then reextract and reanalyze the sample. If the reextraction and reanalysis solve the problem, then the problem was in the analyst's control. Therefore, only submit data from the extraction and analysis with surrogate spike recoveries within the control windows. This will be considered the initial analysis and will be reported as such on all data deliverables.

5.3.4 If the reextraction and reanalysis of the sample do not solve the problem (i.e., surrogate recoveries are outside the control windows for both analyses), then submit the surrogate spike recovery data and the sample data from both analyses. Distinguish between the initial analysis and the reanalysis on all data deliverables.

5.4 Documentation

The laboratory will report surrogate recovery data for the following.

5.4.1 Method blank analysis

5.4.2 Sample analysis

5.4.3 Matrix spike/matrix spike duplicate analysis

5.4.4 All sample reanalyses that substantiate a matrix effect

6. Matrix spike/matrix spike duplicate (MS/MSD) analysis

6.1 MS/MSD frequency of analysis

A matrix spike and matrix spike duplicate must be performed for each group of samples of a similar matrix, once each case of field samples has been received, or each 20 field samples in a case have been analyzed, whichever is more frequent.

6.2 Use the compounds listed in Attachment 10 to prepare matrix spiking solutions. Optional dilution steps must be accounted for when calculating the percent recovery of the matrix spike and matrix spike duplicate samples.

Note: Samples requiring optional dilutions and chosen as the MS/MSD samples must be analyzed at the same dilution as the original unspiked sample.

6.3 Individual component recoveries of the matrix spike are calculated using the following Equation.

$$\text{Matrix spike percent recovery} = \frac{SSR - SR}{SA} \times 100$$

Where:

SSR = Spike sample results.
SR = Sample result.
SA = Spike added from spiking mix.

6.4 Relative percent difference (RPD)

The analyst is required to calculate the RPD between the matrix spike and the matrix spike duplicate. RPDs for each component is calculated using the following Equation.

$$RPD = \frac{D_1 - D_2}{(D_1 + D_2)/2} \times 100$$

Where:

RPD = Relative percent difference
D_1 = First sample value
D_2 = Second sample value (duplicate)

6.5 The matrix spike results (concentrations) for nonspiked, semivolatile TCL compounds are reported, and the matrix spike percent recoveries are summarized (Attachment 11).

APPARATUS:

- Soxhlet extractor, 47 mm i.d. x 170 mm length, Pyrex 3740 L.
- Drying column, 19 mm I.D. chromatographic column with coarse frit. Substitution of a small pad of Pyrex glass wool for the frit will prevent cross-contamination of sample extracts.

- Chromatographic column, glass, 1 cm i.d. x 25 cm length, with 250 mL reservoir, sintered glass frit, and Teflon stopcock
- Concentrator tube, Kuderna-Danish (K-D), 10 mL, graduated, Kontes K-570050-1025. The calibration must be checked at the volumes employed in the test. A ground glass stopper is used to prevent evaporation of extracts.
- Evaporative flask, KD, 500 mL, Kontes K-570001 0500. Attach the flask to the concentrator tube with springs.
- Snyder column, KD, three-ball macro, Kontes K503000 0121
- Snyder column, KD, two-ball micro, Kontes K569001 0219
- Beaker, 250 mL
- Vials, amber glass, 2-mL capacity, with Teflon-lined screw caps.
- Erlenmeyer flask, 500 mL, with ground glass joint 24/40.
- Silicon carbide boiling chips, 10/40 mesh. Heat the chips to 400°C for 30 minutes or soxhlet extract them with methylene chloride.
- Water bath, heated, with concentric ring cover, capable of temperature control (±2°C). The bath should be used in a hood.
- Balance, analytical, capable of accurately weighing ±0.0001 g
- Nitrogen evaporation device equipped with a water bath that can be maintained at 35 to 40°C, N-Evap, Organomation Associates, Inc.
- Gas chromatograph, analytical system complete with a temperature programmable gas chromatograph suitable for splitless injection and all required accessories including syringes, analytical columns, and gases
- Column, 30 m x 0.25 mm (or 0.32 mm), bonded-phase silicone-coated fused silica capillary column, J&W Scientific DB-5. A film thickness of 1.0-micron is recommended because of its larger capacity. A film thickness of 0.25-microns may be used.
- Glass chromatographic column for alumina, 6 mL, 150 mm x 8 mm i.d., column Kontes K-420155 or 5-mL serological pipettes plugged with a small piece of Pyrex glass wool or polyethylene porous disk, Kontes K-420162.
- Pyrex glass wool, prerinsed with appropriate solvents to ensure its cleanliness.

- Mass spectrometer, capable of scanning from 35 to 500 amu every 1 second or less, using 70 volts (nominal) electron energy in the electron impact ionization mode and producing a mass spectrum that meets all required criteria when 50 ng of decafluorotriphenylphosphine (DFTPP) is injected through the gas chromatograph inlet.

  Note: DFTPP criteria must be met before any sample extracts are analyzed.

- Data system: A computer system must be interfaced to the mass spectrometer that allows the continuous acquisition and storage on machine-readable media of all mass spectra obtained throughout the duration of the chromatographic program. The computer must have software that can search any GC/MS data file for ions of a specific mass and plot such ion abundances versus time or scan number. This type of plot is defined as an extracted ion current profile (EICP). Software must also be available that allows integrating the abundance in any EICP between specified time or scan number limits.

Note: Equivalent equipment may be substituted.

REAGENTS:

- Reagent water (Water in which an interferant is not observed at or above the method detection limit of each parameter of interest.)

- Sodium hydroxide solution, 10N. Dissolve 40 g of sodium hydroxide in reagent water and dilute it to 100 mL.

- Sodium thiosulfate, ACS, granular

- Sulfuric acid solution (1:1). Slowly add 50 mL of sulfuric acid, (specific gravity 1.84) to 50 mL of reagent water.

- Ethanol, 200 proof

- Acetone, hexane, methanol, and methylene chloride, pesticide quality

- 2% Ethanol/methylene chloride. Add 20 mL of ethanol to 980 mL of methylene chloride.

- Sodium sulfate, ACS, powdered, Baker anhydrous powder, Catalog No. 73898. Purify the sodium sulfate by heating it at 400°C for 4 hours in a shallow tray, cool it in a desiccator, and store it in a glass bottle.

- Florisil, pesticide residue grade. Activate in an oven at 120°C overnight.

- Surrogate standards, phenol-d_6, 2,4,6-tribromophenol, 2-fluorophenol, nitrobenzene-d_5, terphenyl-d_{14}, and 2-fluorobiphenyl. These are added to all samples and calibration solutions.

- Surrogate standard spiking solution, containing the base/neutral compounds at a concentration of 100 µg/mL and the acid compounds at 200 µg/mL. Store the spiking solutions at 4°C ±2°C in Teflon-sealed containers. The solutions should be checked frequently for stability. These solutions must be replaced after at least 12 months, if comparison with quality control check samples indicates a problem.

- Base/neutral/acid (BNA) Matrix standard spiking solution consisting of:

Base/Neutrals	Acids
1,2,4-trichlorobenzene	pentachlorophenol
acenaphthene	phenol
2,4-dinitrotoluene	2-chlorophenol
pyrene	4-chloro-3-methylphenol
N-nitroso-di-n-propylamine	4-nitrophenol
1,4-dichlorobenzene	

- Prepare the matrix spiking solution to contain each of the above compounds in methanol, with the base/neutrals at 100 µg/1.0 mL and the acid compounds at 200 µg/mL.

- Internal standards, 1,4-dichlorobenzene-d_4, naphthalene-d_8, acenaphthene-d_{10}, phenanthrene-d_{10}, chrysene-d_{12}, perylene-d_{12}. An internal standard solution can be prepared by dissolving 200 mg of each compound in 50 mL of methylene chloride. It may be necessary to use 5 to 10% benzene or toluene in this solution and a few minutes of ultrasonic mixing in order to dissolve all the constituents. The resulting solution will contain each standard at a concentration of 4,000 ng/µL. A 10-µL portion of this solution should be added to each 1 mL of sample extract (5 µL to 0.5 mL). This will yield a concentration of 40 ng/µL of each constituent.

- Calibration standards prepared at 20, 50, 80, 120, and 160 ng/µL. Each calibration standard should contain each compound of interest and each surrogate standard.

  Note: Great care must be taken to maintain the integrity of all standard solutions. Store all standard solutions at -10° to -20°C in screw-cap amber bottles with Teflon-liners. Fresh standards should be prepared at least every 12 months. The continuing calibration standard should be prepared weekly and stored at 4°C ±2°C.

Note: Equivalent reagents may be substituted.

PROCEDURE:

1. Sample storage and holding times

   1.1 Sample storage

   The samples must be protected from light and frozen at -20°C from the time of receipt until extraction and analysis.

   1.2 Holding times

   There are no holding times established for fish tissue samples.

2. Sample extraction

   2.1 Attach Soxhlet extractors (47 mm i.d. x 170 mm length) to 500-mL Erlenmeyer flasks with ground glass joints.

   2.2 Add two plugs of glass wool to each extractor, one to cover the bottom to prevent the sample from entering the solvent return arm and the other to cover the top of the sample.

   2.3 Add 300 mL of glass distilled methylene chloride to the Erlenmeyer flask, along with about five boiling chips. Attach the Erlenmeyer to the Soxhlet extractor.

   2.4 Attach the extractors to the condensers in the fume hood.

   2.5 Adjust the temperature so the extractors cycle at a rate of 12 to 15 cycles per hour.

   2.6 Allow the extractors to rinse for 4 hours, then shut off the heaters and allow them to cool.

   2.7 Remove the condensers and drain all the solvent remaining in the extractors into the Erlenmeyer flask.

   2.8 Discard the solvent and rinse the Erlenmeyer flask with two additional 10-mL portions of methylene chloride.

   2.9 The extractors are now ready for the samples.

   2.10 Weigh 30 g of sodium sulfate or the control matrix into a 250-mL beaker; this will represent the method blank.

   2.11 Weigh two 30 portions of the matrix spike and matrix spike duplicates into 250-mL beakers; these will be used for the matrix spikes.

2.12 Weigh 30-g samples into 250-mL beakers.

2.13 Add 60 g of anhydrous sodium sulfate to each beaker and mix the samples thoroughly, using a stainless steel spatula. More sodium sulfate may be necessary (when a sufficient amount has been added, the sample will appear granular).

2.14 Place the beakers in a fume hood and let them dry for at least 3 hours, stirring occasionally.

2.15 Add 2.0 mL of surrogate spiking solution to each beaker.

2.16 Add 2.0 mL of the matrix spiking solution to each 30-g portion of the sample that was selected as the matrix spike and matrix spike duplicate.

2.17 Remove the glass wool plug from the Soxhlet extractors that have been prerinsed.

2.18 Transfer the entire sample to the extractor and place the glass wool plug on top.

Note: The sample level in the extractor should not exceed the top of the solvent return arm; this will keep the entire sample immersed in solvent during the extraction process.

2.19 Add 150 mL of methylene chloride to the mixing beaker, swirl it and add the solvent to the respective Erlenmeyer flask along with about five boiling chips.

2.20 Attach the Soxhlet extractor to the Erlenmeyer flask.

2.21 Add 200 mL of glass-distilled methylene chloride to each Soxhlet extractor.

2.22 Attach the condensers and set the temperature so that the extractors cycle at a rate of 12 to 15 cycles per hour.

2.23 Let the extractors cycle for 16 hours.

2.24 After 16 hours, shut off the heating elements and allow the samples to cool.

2.25 Drain all of the solvent remaining in the extractor into the Erlenmeyer flask.

2.26 Rinse the extractor with about 50 mL of methylene chloride and drain it into the collection Erlenmeyer flask.

2.27 Assemble a K-D concentrator by attaching a 10-mL concentrator tube to a 500-mL evaporative flask. Other concentration devices or techniques may be used in place of the K-D if equivalency is demonstrated for all compounds listed in this method.

2.28 Pour the extracts through powder funnels containing Whatman #4 filter paper and collect the extracts in the K-D concentrators. Rinse the Erlenmeyer flasks and powder funnels with 20 to 30 mL of methylene chloride to complete the quantitative transfer.

2.29 Add one or two clean boiling chips to the evaporative flasks and attach a three-ball Snyder column. Prewet the Snyder columns by adding about 1 mL of methylene chloride to the top. Place the K-D apparatus on a hot water bath (80° to 90°C) so that the concentrator tube is partially immersed in the hot water and the entire lower rounded surface of the flask is bathed with hot vapor. Adjust the vertical position of the apparatuses and the water temperature, as required, to complete the concentration in 10 to 15 minutes. At the proper rate of distillation, the balls of the column will actively chatter but the chambers will not flood with condensed solvent. When the apparent volume of liquid reaches 5 to 10 mL, remove the K-D apparatus. Allow it to drain and cool for at least 10 minutes.

2.30 Adjust the volume to 10 mL using methylene chloride.

3. Lipid determination

3.1 Set up a log book page to include the following information.

3.1.1 Pan identification number

3.1.2 Sample identification number

3.1.3 Initial pan weight

3.1.4 Final pan and sample weight

3.1.5 Weight of lipids

3.1.6 Percent lipids

3.2 Weigh the aluminum pans on an analytical balance and record weights to four decimal places in a log book.

3.3 Using a disposal pipette, aliquot 1.0 mL of the sample extracts into the pans. record the HWI number in the log book with the corresponding pan identification and weight.

3.4 Place the pans in a hood and loosely cover them with foil (leave the foil loose enough to allow adequate air flow over the samples).

3.5 Let the samples dry overnight (approximately 15 hours).

3.6 Remove the samples from the hood, weigh them on an analytical balance, and record the weight to four decimal places in the log book.

3.7 Determine the weight of lipid as follows.

Weight of lipids = Final pan and sample weight - Initial pan weight.

3.8 The weight of lipid in the 1.0-mL aliquot is used to determine the number of loops required for the GPC cleanup. The maximum weight of lipid that should be loaded on the column at one time is 1.0 g. This means that the maximum extract concentration of lipids would be equivalent to 0.2 g/mL (e.g., 0.2 g/mL using a 5.0-mL sample loop would introduce 1.0 g of lipid to the column).

3.9 Record the weight of lipid in the log book.

3.10 Calculate the percent lipid as follows.

$$\% \text{ Lipid} = \frac{\text{Total weight of lipid in the (10 mL) extract}}{\text{Total weight of sample extracted}} \times 100\%$$

3.11 Record the percent lipid information in the log book.

4. GPC cleanup of extracts

4.1 The remaining 9-mL extract should have the lipids removed by gel permeation chromatography (OP-6004-36).

4.2 Optional: In cases where there is only sufficient sample to be extracted once, remove 4 mL of the 9 mL of remaining extract and save it in case a problem develops during the processing. Dilute the other 5 mL to 10 mL in methylene chloride before loading it onto the GPC.

5. Florisil cleanup of extracts

5.1 Quantitatively transfer the GPC eluent into a 500-mL K-D flask with a 10-mL concentrator tube attached.

5.2 Add a boiling chip and attach a three-ball Snyder column to the K-D apparatus.

5.3 Place the K-D apparatus on a hot water bath and evaporate the methylene chloride down to about 10 mL. Do not allow the sample to go dry!

5.4 Remove the K-D apparatus from the hot water bath. Remove the Snyder column and add about 60 mL of hexane and another boiling chip to the K-D flask. Mix the solvents by tilting the K-D apparatus.

5.5 Put the Snyder column back on the K-D flask, put the K-D apparatus back on the water bath and evaporate the solvent down to about 2 mL.

5.6 Allow the K-D flask to sit and cool for at least 10 minutes.

5.7 Remove the Snyder column and rinse the K-D flask with about 2 mL of hexane.

5.8 Remove the K-D flask from the concentrator tube.

5.9 Place the concentrator tube under a gentle stream of nitrogen and take the sample to a volume of 5 mL.

5.10 The sample may now be cleaned using Florisil.

5.10.1 Place 7 g of the activated Florisil in the column and tap gently to settle the adsorbent bed.

5.10.2 Wash the Florisil by allowing approximately 30 mL MeCl2/EtOH to pass through the column. Close the stopcock when the solvent level is approximately 1 cm above the adsorbent bed. Discard the eluant.

5.10.3 Use the entire sample extract from GPC. Reduce the sample extract volume to about 2 mL and transfer it quantitatively to the column with three 2-mL portions of MeCl2. Place a receiving flask under the column, open the stopcock and allow the extract to percolate through the column. As the solvent level approaches the level of the adsorbent bed, rinse down the sides of the column with about 2 mL of MeCl2 and allow this rinsing to pass onto the column until the solvent level approaches the top of the Florisil bed. Elute the column with 150 mL of the 2% ethanol in methylene chloride solvent mixture.

5.11 Concentrate the eluant to about 4 mL in a KD tube. Concentrate the extract further to just under 1 mL with a gentle stream of nitrogen gas.

5.12 Add 10 µL of BNA internal standard mixture.

5.13 Adjust the final volume to 1.0 mL with methylene chloride.

5.14 Transfer the extract to a GC auto-injection vial.

5.15 Analyze by GC/MS.

6. Calibration

6.1 Each GC/MS system must have the hardware tuned to meet the criteria listed in Attachment 3 for a 50-ng injection of DFTPP. No sample analyses can begin until all these criteria are met. These criteria must be demonstrated each 12-hour shift. DFTPP has to be injected to meet this criterion. Post-acquisition manipulation of abundances is not acceptable.

6.2 The internal standards should permit most components of interest in a chromatogram to have retention times of 0.80 to 1.20 relative to the internal standards. Use the base peak ion from the specific internal standard as the primary ion for quantification (Attachment 5). If interferences are noted, use the next most intense ion as the secondary ion (e.g., for 1,4-dichlorobenzene-d_4 use m/z 152 for quantification)

6.3 The internal standards are added to all calibration standards and all sample extracts just before analysis by GC/MS. A 10-µL aliquot of the internal standard solution should be added to a 1-mL aliquot of calibration standards.

6.4 Analyze 1 µL of each calibration standard and tabulate the area of the primary characteristic ion against concentration for each compound, including the surrogate compounds. Calculate RRFs for each compound using the following:

$$RRF = \frac{A_x}{A_{is}} \times \frac{C_{is}}{C_x}$$

Where:

A_x = Area of the characteristic ion for the compound to be measured.

A_{is} = Area of the characteristic ion for the specific internal standard from Attachment 5.

C_{is} = Concentration of the internal standard (ng/µL).

C^x = Concentration of the compound to be measured (ng/µL).

6.4.1 The average RRF should be calculated for all compounds. A system performance check must be made before this calibration curve is used. Four compounds (the system performance check compounds) are checked for a minimum average RRF. These compounds (the SPCCs) are N-nitroso-di-n-propylamine, hexachlorocyclopentadiene, 2,4-dinitrophenol, and 4-nitrophenol.

6.4.2 A % RSD is calculated for 11 compounds labeled the CCCs on Form VI SV and in Attachment 7. A maximum %RSD is also specified for these compounds. These criteria must be met for the calibration curve to be valid.

6.5 A check of the calibration curve must be performed once every 12 hours during analysis. These criteria are described in detail 5 in the Quality Assurance section. The minimum RRF for the system performance check compounds must checked. If this criterion is met, the RRFs of all compounds are calculated. A percent difference of the daily (12-hour) RRF compared with the average RRF from the initial curve is calculated. A maximum percent difference of 30% is allowed for each compound flagged as "CCC." Only after both these criteria are met can sample analysis begin.

6.6 Internal standard responses and retention times in all standards must be evaluated during or immediately after data acquisition. If the retention time for any internal standard changes by more than 30 seconds from the latest daily (12-hour) calibration standard, the chromatographic system must be inspected for malfunctions and corrections must be made as required. The EICP of the internal standards must be monitored and evaluated for each standard. If the EICP area for any internal standard changes by more than a factor of two (-50% to +100%), the mass spectrometric system must be inspected for malfunction and corrections must be made as appropriate. When corrections are made, the samples analyzed while the system was malfunctioning must be reanalyzed.

7. GCMS analysis

7.1 The following instrumental parameters are required for all performance tests and for all sample analyses.

Electron energy - 70 volts (nominal)
Mass range - 35 to 500 amu
Scan time - not to exceed 1 second/scan

7.2 Internal standard solution (10 µL) is added to each sample extract. Analyze the extract by GC/MS using a bonded-phase silicone-coated fused silica capillary column. The recommended GC operating conditions to be used are as follows:

Initial column temperature hold: 40°C for 4 minutes
Column temperature program: 40° to 270°C at 10°C/minute
Final column temperature hold: 270°C for 10 minutes
Injector temperature: 250° to 300°C
Transfer line temperature: 250° to 300°C
Source temperature: According to manufacturer's specifications
Injector: Grob-type, splitless
Sample volume: 1 µL
Carrier gas: Helium at 30 cm^3/second

8. Qualitative analysis

8.1 The compounds listed in the TCL will be identified by an analyst who is competent in the interpretation of mass spectra by comparison of the sample mass spectrum with the mass spectrum of a standard of the suspected compound. Two criteria must be satisfied to verify the identifications: elution of the sample component at the GC relative retention time as the standard component, and correspondence of the sample component and standard component mass spectra.

8.1.1 For establishing correspondence of the GC relative retention time (RRT), the sample component RRT must compare within ±0.06 RRT units of the RRT of the standard component. For reference, the standard must be run on the same shift as the sample. If co-elution of interfering components prohibits accurate assignment of the sample component RRT from the total ion chromatogram, the RRT should be assigned by using extracted ion current profiles for ions unique to the component of interest.

8.1.2 For comparison of standard and sample component mass spectra, mass spectra obtained on the analyst's GC/MS are required. Once obtained, these standard spectra may be used for identification purposes, <u>only</u> if the GC/MS meets the DFTPP daily tuning requirements. These standard spectra may be obtained from the run used to obtain reference RRTs.

8.1.3 The requirements for qualitative verification by comparison of mass spectra are as follows:

8.1.3.1 All ions present in the standard mass spectra at a relative intensity greater than 10% (most abundant ion in the spectrum equals 100%) <u>must</u> be present in the sample spectrum.

8.1.3.2 The relative intensities of ions specified in Step 5.1.3.1 must agree within ±20% between the standard and sample spectra. (e.g., for an ion with an abundance of 50% in the standard spectra, the corresponding sample ion abundance must be between 30% and 70%).

8.1.3.3 Ions greater than 10% in the sample spectrum, but not present in the standard spectrum, must be considered and accounted for by the analyst making the comparison. The verification process should favor false positives. All compounds meeting the identification criteria must be reported with their spectra. For all compounds below the method detection limit (MDL) report the actual value followed by "J" (e.g., "3J").

8.1.4 If a compound cannot be verified by all the criteria in Step 5.1.3, but the identification is correct in the technical judgement of the mass spectral interpretation specialist, then the contractor will report that identifications and proceed with quantification (Calculations section).

8.2 A library search will be executed for non-TCL sample components for the purpose of tentative identification. For this purpose, the 1985 release of the National Bureau of Standards (NBS) Mass Spectral Library (or a more recent release), containing 42,261 spectra, will be used.

8.2.1 Up to 20 nonsurrogate organic compounds of greatest apparent concentration not listed for the combined BNA fraction will tentatively be identified via a forward search of the NBS mass spectral library. (Substances with responses less than 10% of the nearest internal standard are not required to be searched in this fashion.) Only after visual comparison of sample spectra with the nearest library searches will the mass spectral interpretation specialist assign a tentative identification.

8.2.2 Guidelines for making identification.

8.2.2.1 Relative intensities of major ions in the reference spectrum (ions greater than 10% of the most abundant ion) should be present in the sample spectrum.

8.2.2.2 The relative intensities of the major ions should agree within ±20% (e.g., for an ion with an abundance of 50% in the standard spectra, the corresponding sample ion abundance must be between 30 and 70%).

8.2.2.3 Molecular ions present in reference spectrum should be present in sample spectrum.

8.2.2.4 Ions present in the sample spectrum, but not in the reference spectrum, should be reviewed for possible background contamination or presence of coeluting compounds.

8.2.2.5 Ions present in the reference spectrum but not in the sample spectrum, should be reviewed for possible subtraction from the sample spectrum because of background contamination or coeluting compounds.

8.2.3 If in the technical judgement of the mass spectral interpretation specialist no valid, tentative identification can be made, the compound should be reported as unknown. The mass spectral specialist should give additional classification of the unknown compound, if possible (e.g., unknown phthalate, unknown hydrocarbon, unknown acid type, unknown chlorinated compound). If probable molecular weights can be distinguished, include them.

CALCULATIONS:

1. Identified TCL components will be quantified by the internal standard method. The internal standard used will be the one with a retention time nearest that of a given analyte. The EICP area of characteristic ions of analytes listed in Attachments 4 and 5 are used.

   Internal standard responses and retention times in all samples must be evaluated during or immediately after data acquisition. If the retention time for any internal standard changes by more than 30 seconds from the latest daily (12-hour) calibration standard, the chromatographic system must be inspected for malfunctions and corrections must be made as required. The EICP of the internal standards must be monitored and evaluated for each sample, blank, matrix spike, and matrix spike duplicate. If the EICP area for any internal standard changes by more than a factor of two (-50% to +100%), the mass spectrometric system must be inspected for malfunction and corrections must be made as appropriate. When corrections are made, the samples analyzed while the system was malfunctioning must be reanalyzed.

2. The RRF from the daily standard analysis is used to calculate the concentration in the sample. Secondary ions may be used if interferences are present. The area of a secondary ion cannot be substituted for the area of a primary ion unless an RRF is calculated using the secondary ion. When TCL compounds are below quantitation limits, but the spectra meet the identification criteria, report the concentration with a "J."

   Calculate the concentration in the sample using the RRF, as determined in Step 4.3 of the Procedure section and the following equation.

$$\text{Concentration } \mu g/kg = \frac{(A_x)(I_s)(V_t)}{(A_{is})(RRF)(W_s)(V_i)(D)}$$

   A_x = Area of the characteristic ion for the compound to be measured

   A_{is} = Area of the characteristic ion for the internal standard

   I_s = Amount of internal standard injected in nanograms (ng)

   W_s = Weight of sample extracted (grams)

   V_i = Volume of extract injected (μL)

   V_t = Volume of total extract (the volume is 2000 μL if GPC cleanup is used).

   $D = \frac{100 - \%\ \text{moisture}}{100}$

3. An estimated concentration for tentatively identified non-TCL components will be quantified by the internal standard method. For quantification, the nearest internal standard <u>free of interferences</u> will be used.

   The formula for calculating concentrations is the same as that in Step 20. Total area counts (or peak heights) from the total ion chromatograms are to be used for both the compound to be measured and the internal standard. An RRF of 1 is to be assumed. The value from this quantitation will be qualified as estimated. This estimated concentration should be calculated for all tentatively identified compounds, as well as those identified as unknowns.

4. Calculate surrogate standard recovery on all samples, blanks, and spikes. Determine if the recovery is within the limits and report it.

   4.1 If the recovery is not within the limits (i.e., if two surrogates from either base/neutral or acid fractions are out of limits, <u>or</u> if recovery of any <u>one</u> surrogate in either fraction is below 10%), the following steps are required.

4.1.1 Check to be sure there are no errors in calculations, surrogate solutions, and internal standards.

4.1.2 Check instrument performance.

4.1.3 Reanalyze the sample if none of the above reveal a problem.

4.2 If none of the steps above solve the problem, then reextract and reanalyze the sample. If the reextraction and reanalysis of the sample solves the problem, then the problem was within the laboratory's control. Therefore, only submit data from the analysis with surrogate spike recoveries within the control windows. This will be considered the initial analysis and will be reported as such.

4.3 If the reextraction and reanalysis of the samples does not solve the problem (i.e., the surrogate recoveries are outside the control limits for both analyses), then submit the surrogate spike recovery data and the sample analysis data from analysis of both sample extracts. Distinguish between the initial analysis and the reanalysis on all data deliverables.

4.4 If the sample with surrogate recoveries outside the limits is the sample used for the matrix spike and matrix spike duplicate, and the surrogate recoveries of the matrix spike and the matrix spike duplicates show the same pattern (i.e., outside the limits), then the sample, matrix spike, and matrix spike duplicate do not require reanalysis. Document in the narrative the similarity in surrogate recoveries.

TARGET COMPOUND LIST

CAS No.	Compound	Target Detection Limits (µg/kg)
108-95-2	Phenol	660
111-44-4	bis (2-Chloroethyl) Ether	660
95-57-8	2-Chlorophenol	660
541-73-1	1,3-Dichlorobenzene	660
106-46-7	1,4-Dichlorobenzene	660
100-51-6	Benzyl Alcohol	660
95-50-1	1,2-Dichlorobenzene	660
95-48-1	2-Methylphenol	660
39638-32-9	bis(2-Chloroisopropyl)Ether	660
106-44-5	4-Methylphenol	660
621-64-7	N-Nitroso-Di-n-Propylamine	660
67-72-1	Hexachloroethane	660
98-95-3	Nitrobenzene	660
78-59-1	Isophorone	660
88-75-5	2-Nitrophenol	660
105-67-9	2,4-Dimethylphenol	660
65-85-0	Benzoic Acid	3,200
111-91-1	bis(2-Chloroethoxy)Methane	660
120-83-2	2,4-Dichlorophenol	660
120-82-1	1,2,4-Trichlorobenzene	660
91-20-3	Naphthalene	660
106-47-8	4-Chloroaniline	660
87-68-3	Hexachlorobutadiene	660
59-50-7	4-Chloro-3-methylphenol	660
91-57-6	2-Methylnaphthalene	660
77-47-4	Hexachlorocyclopentadiene	660
88-06-2	2,4,6-Trichlorophenol	660
95-95-4	2,4,5-Trichlorophenol	3,200
91-58-7	2-Chloronaphthalene	660
88-74-4	2-Nitroaniline	3,200
131-11-3	Dimethyl Phthalate	660
208-96-8	Acenaphthylene	660
606-20-2	2,6-Dinitrotoluene	660
99-09-2	3-Nitroaniline	3,200
83-32-9	Acenaphthene	660
51-28-5	2,4-Dinitrophenol	3,200
100-02-7	4-Nitrophenol	3,200
132-64-9	Dibenzofuran	660
121-14-2	2,4-Dinitrotoluene	660
84-66-2	Diethylphthalate	660
7005-72-3	4-Chlorophenyl-Phenylether	660
86-73-7	Fluorene	660
100-10-6	4-Nitroaniline	3,200
534-52-1	4,6-Dinitro-2-Methylphenol	3,200
86-30-6	N-Nitrosodiphenylamine (1)	660

TARGET COMPOUND LIST

CAS No.	Compound	Target Detection Limits (µg/kg)
101-55-3	4-Bromophenyl-Phenylether	660
118-74-1	Hexachlorobenzene	660
87-86-5	Pentachlorophenol	3,200
85-01-8	Phenanthrene	660
120-12-7	Anthracene	660
84-74-2	Di-n-butylphthalate	660
206-44-0	Fluoranthene	660
129-00-0	Pyrene	660
85-68-7	Butylbenzylphthalate	660
91-94-1	3,3'-Dichlorobenzidine	1,300
56-55-3	Benzo(a)anthracene	660
218-01-9	Chrysene	660
117-81-7	bis(2-Ethylhexyl)phthalate	660
117-84-0	Di-n-octylphthalate	660
205-99-2	Benzo(b)fluoranthene	660
207-08-9	Benzo(k)fluoranthene	660
50-32-8	Benzo(a)pyrene	660
193-39-5	Indeno(1,2,3-cd)Pyrene	660
53-70-3	Dibenz(a,h)Anthracene	660
191-24-2	Benzo(g,h,i)perylene	660

SEMIVOLATILE HAZARDOUS SUBSTANCE LIST COMPOUNDS
FISH QUALITY CONTROL SUMMARY

Compound	Spike Added (ppm)	Determinant Number of Samples	Average % Recovered	Standard Deviation
Phenol	1.67	15	70.5	13.97
bi(2-Chloroethyl)ether	1.67	14	55.6	11.15
2-Chlorophenol	1.67	16	60.6	13.83
1,3-Dichlorobenzene	1.67	16	35.8	18.54
1,4-Dichlorobenzene	1.67	16	36.3	17.77
Benzyl Alcohol	1.67	14	57.7	23.09
1,2-Dichlorobenzene	1.67	16	40.2	19.65
2-Methylphenol	1.67	16	63.2	20.35
bis(2-Chloroisopropyl)ether	1.67	16	55.1	19.04
4-Methylphenol	1.67	16	69.1	12.37
N-Nitroso-di-n-propylamine	1.67	14	68.9	21.94
Hexachloroethane	1.67	16	35.1	19.31
Nitrobenzene	1.67	16	61.9	20.32
Isophorone	1.67	14	75.4	19.96
2-Nitrophenol	1.67	15	59.9	18.81
2,4-Dimethylphenol	1.67	14	67.4	20.98
Benzoic Acid	8.33	16	0.0	0.00
bis(2-Chloroethoxy)methane	1.67	16	73.4	18.48
2,4-Dichlorophenol	1.67	16	72.6	13.04
1,2,4-Trichlorobenzene	1.67	15	60.4	19.87
Naphthalene	1.67	16	65.7	18.21
4-Chloroaniline	1.67	16	20.9	15.30
Hexachlorobutadiene	1.67	15	52.0	17.13
4-Chloro-3-methylphenol	1.67	16	72.7	15.17
2-Methylnaphthalene	1.67	16	71.3	14.36
Hexachlorocyclopentadiene	1.67	15	3.1	5.71
2,4,6-Trichlorophenol	1.67	16	82.2	17.46
2,4,5-Trichlorophenol	8.33	16	74.6	14.22
2-Chloronaphthalene	1.67	14	77.1	15.14
2-Nitroaniline	8.33	15	84.7	15.57
Dimethyl Phthalate	1.67	14	90.9	21.83
Acenaphthylene	1.67	16	87.1	19.27
3-Nitroaniline	8.33	15	55.7	24.17
Acenaphthene	1.67	16	85.5	17.59
2,4-Dinitrophenol	8.33	16	6.1	16.09
4-Nitrophenol	8.33	14	72.4	28.58
Dibenzofuran	1.67	16	86.2	17.44
2,4-Dinitrotoluene	1.67	16	85.1	20.32
2,6-Dinitrotoluene	1.67	16	86.8	18.14
Diethylphthalate	1.67	14	89.6	27.49
4-Chlorophenyl-phenylether	1.67	16	83.0	17.39
Fluorene	1.67	16	83.8	17.17
4-Nitroaniline	8.33	16	29.0	20.54
4,6-Dinitro-2-methylphenol	8.33	15	21.5	28.12

SEMIVOLATILE HAZARDOUS SUBSTANCE LIST COMPOUNDS
FISH QUALITY CONTROL SUMMARY

Compound	Spike Added (ppm)	Determinant Number of Samples	Average % Recovered	Standard Deviation
N-Nitrosodiphenylamine	1.67	16	75.4	19.38
4-Bromophenyl-phenylether	1.67	16	86.7	17.53
Hexachlorobenzene	1.67	15	93.3	17.86
Pentachlorophenol	8.33	14	75.4	29.38
Phenanthrene	1.67	14	94.1	18.20
Anthracene	1.67	16	86.5	19.24
Di-n-butylphthalate	1.67	13	102.2	30.33
Fluoranthene	1.67	14	100.1	21.28
Pyrene	1.67	14	79.2	19.82
Butylbenzyphthalate	1.67	14	97.0	29.70
3,3-Dichlorobenzidine	3.33	15	7.8	15.13
Benzo(a)anthracene	1.67	14	84.1	20.05
bis(2-Ethylhexyl)phthalate	1.67	13	88.7	39.12
Chrysene	1.67	16	86.8	20.77
Di-n-octyl Phthalate	1.67	13	65.9	33.01
Benzo(b)Fluoranthene	1.67	14	85.2	23.90
Benzo(k)Fluoranthene	1.67	14	90.6	21.89
Benzo(a)Purene	1.67	15	77.2	19.84
Indeno(1,2,3-cd)pyrene	1.67	13	74.4	38.30
Dibenzo(a,h)anthracene	1.67	13	81.8	36.12
Benzo(g,h,i)perylene	1.67	13	63.9	32.40

DFTPP KEY IONS AND ION ABUNDANCE CRITERIA

Mass	Ion Abundance Criteria
51	30.0% to 60.0% of mass 198
68	Less than 2.0% of mass 69
70	Less than 2.0% of mass 69
127	40.0% to 60.0% of mass 198
197	Less than 1.0% of mass 198
198	Base peak, 100% relative abundance
199	5.0% to 9.0% of mass 198
275	10.0% to 30.0% of mass 198
365	Greater than 1.00% of mass 198
441	Present but less than mass 443
442	Greater than 40.0% of mass 198
443	17.0% to 23.0% of mass 442

CHARACTERISTIC IONS FOR SEMIVOLATILE TCL COMPOUNDS

Parameter	Primary Ion	Secondary Ion(s)
Phenol	94	65, 66
bis(-2-chloroethyl)ether	93	63, 95
2-Chlorophenol	128	64, 130
1,3-Dichlorobenzene	146	148, 113
1,4-Dichlorobenzene	146	148, 113
Benzyl alcohol	108	79, 77
1,2-Dichlorobenzene	146	148,113
2-Methylphenol	108	107
bis(-2-chloroisoprophyl)ether	45	77, 79
4-Methylphenol	108	107
N-Nitroso-di-propylamine	70	42, 101, 130
Hexachloroethane	117	201, 199
Nitrobenzene	77	123, 65
Isophorone	82	95, 138
2-Nitrophenol	139	65, 109
2,4-Dimethylphenol	107	121, 122
Benzoic acid	122	105, 77
bis(-2-chloroethoxy)methane	93	95, 123
2,4-Dichlorophenol	162	164, 98
1,2,4-Trichlorobenzene	180	182, 145
Naphthalene	128	129, 127
4-Chloroaniline	127	129
Hexachlorobutadiene	225	223, 227
4-Chloro-3-methylphenol	107	144, 142
2-Methylnaphthalene	142	141
Hexachlorocyclopentadiene	237	235, 272
2,4,6-Trichlorophenol	196	198,200
2,4,5-Trichlorophenol	196	198,200
2-Chloronaphthalene	162	164, 127
2-Nitroaniline	65	92, 138
Dimethyl phthalate	163	194, 164
Acenaphthylene	152	151, 153
3-Nitroaniline	138	108, 92
Acenaphthene	153	152, 154
2,4-Dinitrophenol	184	63, 154
4-Nitrophenol	109	139, 65
Dibenzofuran	168	139
2,4-Dinitrotoluene	165	63, 182
2,6-Dinitrotoluene	165	89, 121
Diethylphthalate	149	177, 150
4-Chlorophenyl-phenylether	204	206, 141
Fluorene	166	165, 167
4-Nitroaniline	138	92, 108
4,6-Dinitro-2-Methylphenol	198	182, 77
N-Nitrosodiphenylamine	169	168, 167
4-Bromophenyl-phenylether	248	250, 141
Hexachlorobenzene	284	142, 249

CHARACTERISTIC IONS FOR SEMIVOLATILE TCL COMPOUNDS

Parameter	Primary Ion	Secondary Ion(s)
Pentachlorophenol	266	264, 268
Phenanthrene	178	179, 176
Anthracene	178	179, 176
Di-n-Butylphthalate	149	150, 104
Fluoranthene	202	101, 100
Pyrene	202	101, 100
Butylbenzylphthalate	149	91, 206
3,3'-Dichlorobenzidine	252	254, 126
Benzo(a)anthracene	228	229, 226
bis(2-ethylhexyl)phthalate	149	167, 279
Chrysene	228	226, 229
Di-n-octyl phthalate	149	-
Benzo(b)fluoranthene	252	253, 125
Benzo(k)fluoranthene	252	253, 125
Benzo(a)pyrene	252	253, 125
Indeno(1,2,3-cd)pyrene	276	138, 227
Dibenz(a, h)anthracene	278	139, 279
Benzo(g, h, i)perylene	276	138, 277

CHARACTERISTIC IONS FOR SURROGATES AND INTERNAL STANDARDS FOR SEMIVOLATILE COMPOUNDS

Parameter	Primary Ion	Secondary Ion(s)
	Surrogates	
Phenol-d_5	99	42, 71
2-Fluorophenol	112	64
2,4,6-Tribromophenol	330	332, 141
d-5 Nitrobenzene	82	128, 54
2-Fluorobiphenyl	172	171
Terphenyl	244	122, 212
	Internal Standards	
1,4-Dichlorobenzene-d_4	152	115
Naphthalene-d_8	136	68
Acenaphthene-d_{10}	164	162, 160
Phenanthrene-d_{10}	188	94, 80
Chrysene-d_{10}	240	120, 236
Perylene-d_{12}	264	260, 265

Semi-Volatile Internal Standards with Corresponding TCL Analytes Assigned for Quantitation

1,4-Dichlorobenzene-d	Naphthalene-d	Acenaphthene-d	Phenanthrene-d	Chrysene-d	Perylene-d
Phenol	Nitrobenzene	Hexachlorocyclopentadiene	4,6-Dinitro-2-methylphenol	Pyrene	Di-n-octyl phthalate
bis(2-chloroethyl) ether	Isophorone	2,4,6-Trichlorophenol	N-nitrosodiphenylamine	Butylbenzyl phthalate	Benzo(b)fluoranthene
2-Chlorophenol	2-Nitrophenol	2,4,5-Trichlorophenol	1,2-Diphenylhydrazine	3,3-Dichlorobenzidine	Benzo(k)fluoranthene
1,3-Dichlorobenzene	2,4-Dimethylphenol	2-Chloronaphthalene	4-Bromophenyl phenyl ether	Benzo(a)anthracene	Benzo(a)pyrene
1,4-Dichlorobenzene	Benzoic acid	2-Nitroaniline	Hexachlorobenzene	bis(2-ethylhexyl)phthalate	Indeno(1,2,3-c pyrene
Benzyl alcohol	bis(2-Chloroethoxy)methane	Dimethyl phthalate	Pentachlorophenol	Chrysene	Dibenz(a, h) anthracene
1,2-Dichlorobenzene	2,4-Dichlorophenol	Acenaphthylene	Phenanthrene	Terphenyl-d_{14} (surr)	Benzo(g, h, i) perylene
2-Methylphenol	1,2,4-Trichlorobenzene	3-Nitroaniline	Anthracene		
bis(2-Chloroisopropyl)ether	Napthalene	Acenaphthene	Di-n-butyl phthalate		
4-Methylphenol	4-Chloroaniline	2,4-Dinitrophenol	Fluoranthene		
N-nitroso-Di-n-propylamine	Hexachlorobutadiene	4-Nitrophenol			
Hexachloroethane	4-Chloro-3-methylphenol	Dibenzofuran			
2-Fluorophenol (surr)	2-Methylnaphthalene	2,4-Dinitrotoluene			
Phenol-d_6 (surr)	Nitrobenzene-d_5 (surr)	2,6-Dinitrotoluene			
		Diethyl phthalate			
		4-Chlorophenyl-phenyl ether			
		Fluorene			
		4-Nitroaniline			
		2-Fluorobiphenyl (Surr)			
		2,4,6-Tribromophenol			

Calibration Check Compounds

Base/Neutral Fraction	Acid Fraction
Acenaphthene	4-Chloro-3-Methylphenol
1,4-Dichlorobenzene	2,4-Dichlorophenol
Hexachlorobutadiene	2-Nitrophenol
N-Nitroso-di-n-phenylamine	Phenol
Di-n-octylphthalate	Pentachlorophenol
Fluoranthene	2,4,6-Trichlorophenol
Benzo(a)pyrene	

Continuing calibration: 50 ng

Surrogate Spiking Compounds

Compound	Amount in Sample Fraction	Amount in Sample (µg)
Nitrobenzene-d_5	BNA	200
2-Fluorobiphenyl	BNA	200
p-Terphenyl-d_{14}	BNA	200
Phenol-d_5	BNA	400
2-Fluorophenol	BNA	400
2,4,6-Tribromophenol	BNA	400

Required Surrogate Spike Recovery Limits

Fraction	Surrogate Compound	%
BNA	Nitrobenzene-d_5	23-120
BNA	2-Fluorobiphenyl	30-115
BNA	p-Terphenyl-d_{14}	18-137
BNA	Phenol-d_5	24-113
BNA	2-Fluorophenol	25-121
BNA	2,4,6-Tribromophenol	19-122

Matrix Spiking Solutions

Base/Neutrals	Spike Amount (µg)	Acids	Spike Amount (µg)
1,2,4-Trichlorobenzene	200	Pentachlorophenol	400
Acenaphthene	200	Phenol	400
2,4-Dinitrotoluene	200	2-Chlorophenol	400
Pyrene	200	4-Chloro-3-Methylphenol	400
1,4-Dichlorobenzene	200	4-Nitrophenol	400

Matrix Spike Recovery Limits

Fraction	Matrix Spike Compound	(%)	%RSD
BN	1,2,4-Trichlorobenzene	10-120	33
BN	Acenaphthene	29-145	23
BN	2,4-Dinitrotoluene	24-146	24
BN	Pyrene	20-139	25
BN	N-Nitroso-Di-n-Propylamine	10-135	32
BN	1,4-Dichlorobenzene	10-90	49
Acid	Pentachlorophenol	10-164	39
Acid	Phenol	28-112	20
Acid	2-Chlorophenol	19-102	23
Acid	4-Chloro-3-Methylphenol	27-118	21
Acid	4-Nitrophenol	10-158	40