# Prescribed Analytical Procedure (PAP) – Volume 2
# Analyte Group 2: PCB Congeners by Low Resolution Mass Spectrometry and coplanar/mono-*ortho* substituted PCB Congeners by High Resolution Mass Spectrometry in Biological Tissue Samples

## Contents



## Analyte Group 2 Contract Manager

Any issue, problem, or question encountered during the performance of work under Analyte Group 2 must be brought to the attention of the Analyte Group 2 Contract Manager. Jesse Becker in the Division of Fish & Wildlife (DFW) is the current Analyte Group 2 Contract Manager:


Jesse Becker, Research Scientist 2
Division of Fish & Wildlife
New York State Dept. of Environmental Conservation
625 Broadway – 5th Floor
Albany, New York 12233-4756
Voice: (518) 402-8973
FAX: (518) 402-8925
jesse.becker@dec.ny.gov


In addition to the Analyte Group 2 Contract Manager detailed above, all email correspondence related to Analyte Group 2 must be copied to the current NYSDEC Project Manager, as designated by the head of the Environmental Monitoring Section.

## EXHIBIT A – Summary of Requirements

A-1. Overview of Analyte Group: This analyte group involves the analysis of biological tissue samples for polychlorinated biphenyl (PCBs) congeners by low resolution mass spectrometry, with additional resolution and quantification of coplanar/non-*ortho* and mono-*ortho* substituted PCB congeners by high resolution mass spectrometry (i.e., the 12 PCB congeners identified by the World Health Organization [WHO] as "dioxin-like" and included in the Toxic Equivalency Factor [TEF] scheme; Van den Berg et al. 2006); and lipids and moisture.

A-2. Laboratory Qualifications: Laboratories performing work within this analyte group are required to have the equipment, personnel, and knowledge to analyze biological tissue samples for these compounds using methods such as: Method 625 (U.S. EPA 1984) or Method 8270D (U.S. EPA 2014c) for GC/LRMS and Method 1668C (U.S. EPA 2010) for HRGC/HRMS, and gravimetric methods for lipids and moisture. The laboratory must have the ability to perform additional cleanup procedures on tissue extracts to focus HRGC/HRMS analysis on the WHO-TEF congeners. Other analytical methods that produce analytical determinations having similar or better quality and/or lower detection limits may be acceptable to the Department (e.g., full quantification by HRGC/HRMS). If such methods are standard methods, the methods should be cited. If methods other than standard methods are anticipated or proposed, the methods must be submitted, along with primary literature support, to the Department for review and approval at the time of response to the IFB and must be accompanied by proof of method performance (see Exhibit D).

A-3. Staffing: The technical staff of the laboratory performing work within this analyte group must have the qualifications as defined in the Invitation for Bid (IFB)-Article 4(b). This includes a Project Manager, Technical Director, and QA/QC Officer.

A-4. Equipment and Instrumentation: Analytical instruments and all equipment of each type needed for each portion of the analysis. One (1) GC/LRMS instrument capable of meeting or configurable to the instrument specifications of EPA Method 625 or 8270D and one (1) HRGC/HRMS instrument capable of meeting, or configurable to, the instrument specifications of EPA Method 1668C. Each instrument or piece of equipment must meet the specifications needed for the analysis in such a way to meet the Contract Required Quantification Limits (CRQLs) as listed in Exhibit C and the Analyte Group 2 Price Tables (IFB Attachment 1).

A-5. Certification and Licenses: None required.

A-6. Rush Turnaround Samples: There is no requirement for rush turnaround samples for this Analyte Group.

A-7. Sample Identifiers: Samples will be identified by project name and traceable to the individual sample tag sample numbers on the Chain of Custody forms supplied by NYSDEC. Any number of samples can be shipped at a time; however, the vendor will organize individual samples in each shipment into Sample Delivery Groups (SDGs).

SDGs will consist of subsets of twenty (20) individual samples. An SDG can consist of fewer than 20 individual sample numbers if the total number of samples in a shipment is not evenly divisible by 20. Performance evaluation or reference material samples analyzed with an SDG do not contribute to the 20-sample limit.

A-8. Invoicing and Payments

A. Responsible Party: The Analyte Group 2 Contract Manager (Jesse Becker) should be listed as the "Bill to:" or "Invoice to:" party on all invoices generated under Analyte Group 2. The Analyte Group 2 Contract Manager will also serve as the primary point of contact for all issues related to billing and payments.

B. Invoice Distribution: : Upon the completion of the work ordered, invoices must be distributed as electronic copies to both:

1. The Analyte Group 2 Contract Manager (jesse.becker@dec.ny.gov).
2. The Analyte Group 2 Project Manager (to be determined).

C. Invoice Format: The following informational items must be included on all invoice submitted to the Department for payment. All listed items must be included on the front page of the invoice, unless marked with a "*". All "*" items are still required but may be included on an attachment.

1. Invoice number
2. Contract number
3. Contractor EIN
4. Invoice date
5. DEC project name
6. Internal laboratory SDG
7. Date samples were received
8. Analyses ordered
9. Number of samples processes/analyzed
10. Unit price for each analysis line item
11. Total price for each line item
12. Invoice grand total
13. Sample ID's *
14. Analyses ordered for each sample ID *

D. Figure A-8.1 is an example of an acceptable invoice attachment summarizing analyses ordered by sample (Lines 13 & 14 in Item A-8C, above).

E. Invoice Errors and Rejection: If an invoice does not meet any of the requirements herein or of the contract itself, it will be rejected by the Department, with instructions for revision. Revision requests will come from the Analyte Group 2 Contract Manager or designee via email and will reference the invoice in question. Revised invoices must have their "date issued" updated to reflect the date of revision.

Figure A-8.1: Example Invoice Attachment

*Sample Summary Attachment of Invoice Number 123456*

| NYSDEC Sample ID # | Laboratory ID/Job # | Tests Ordered | Date Received | Date Processed |
|---|---|---|---|---|
| 0261063 | NYHR10-001 | lipids, Moisture, PFCs PCNs HBCD, TBBPA | 6/14/2012 | 8/02/2012 |
| 0261064 | NYHR10-002 | | 6/14/2012 | 8/02/2012 |
| 0261065 | NYHR10-003 | | 6/14/2012 | 8/02/2012 |
| 0261066 | NYHR10-004 | | 6/14/2012 | 8/02/2012 |
| 0261067 | NYHR10-005 | | 6/14/2012 | 8/02/2012 |
| 0261068 | NYHR10-006 | | 6/14/2012 | 8/02/2012 |
| 0261069 | NYHR10-007 | | 6/14/2012 | 8/02/2012 |
| 0261070 | NYHR10-008 | | 6/14/2012 | 8/02/2012 |
| 0261071 | NYHR10-009 | | 6/14/2012 | 8/02/2012 |
| 0261072 | NYHR10-010 | | 6/14/2012 | 8/02/2012 |
| 0261073 | NYHR10-011 | | 6/14/2012 | 8/02/2012 |
| 0261074 | NYHR10-012 | | 6/14/2012 | 8/02/2012 |
| 0261075 | NYHR10-013 | | 6/14/2012 | 8/02/2012 |
| 0261076 | NYHR10-014 | | 6/14/2012 | 8/02/2012 |
| 0261077 | NYHR10-015 | | 6/14/2012 | 8/02/2012 |
| 0261078 | NYHR10-016 | | 6/14/2012 | 8/02/2012 |
| 0261079 | NYHR10-017 | | 6/14/2012 | 8/02/2012 |
| 0261080 | NYHR10-018 | | 6/14/2012 | 8/02/2012 |
| 0261081 | NYHR10-019 | | 6/14/2012 | 8/02/2012 |
| 0261082 | NYHR10-020 | | 6/14/2012 | 8/02/2012 |
| HRM198 | NYHR10-021 | | 6/14/2012 | 8/02/2012 |
| HRM199 | NYHR10-022 | | 6/14/2012 | 8/02/2012 |
| HRM200 | NYHR10-023 | | 6/14/2012 | 8/02/2012 |
| HRM201 | NYHR10-024 | | 6/14/2012 | 8/02/2012 |

A-9. Refusal of Work: It is understood that under certain circumstances a laboratory may not be able to complete certain analyses requested by the Department. The Department does not maintain redundant contract services in many cases, and it is also very difficult to obtain services outside of the laboratory contracts. For these reasons, the Department needs to keep refusals of work to a minimum. The refusal clauses below apply to all analysis line items specified in Analyte Group 2.

A. Acceptable Conditions for Work Refusal (see also IFB Article 3(b)(2)):

1. Instrumentation – If the instrumentation required to perform a requested analysis is not available or inoperable, and no redundant instrumentation is available, work may be refused. If the instrument is only temporarily unavailable and applicable holding times and reporting deadlines can be met, samples must be accepted.
2. Conflict of Interest – If the laboratory already has as a client, a party regulated by the Department, and the Department is performing split sampling with that same regulated party, the laboratory must notify the Department upon receipt of such samples. Split samples with an existing client of the laboratory may be refused as the intent of split sampling is to use unique laboratories. See IFB, Article 12 for additional details.
3. Contamination – If the laboratory has a contamination issue within a laboratory area or analytical system, defined by two (2) method blank failures in a 7-day period, the Department must be notified. If the contamination will affect the integrity or accuracy of samples analyzed for the Department, the laboratory must refuse potentially affected samples.

B. Notifications: If the laboratory chooses to exercise any of the above refusal clauses, the Department must be notified immediately to ensure any applicable holding times can be met. Notifications should be made to the sample submitter and the Analyte Group 2 Contract Manager by phone or by email. A follow-up written notice must also be made to the Analyte Group 2 Contract Manager providing a detailed explanation of why the samples were refused.

C. Subcontracting of Refused Work (See also IFB Article 14):

1. Samples meeting refusal criteria – If the laboratory decides to rightfully refuse any samples submitted by the Department, and no redundant contractors are available to the Department in the applicable Analyte Group, the samples must be subcontracted to a third-party laboratory. The cost for said analyses will be the line item price previously specified in the contract. No up-charge for subcontracting shall apply.
2. High-level Samples – The laboratory is not allowed to refuse samples based on a high-level of target or non-target analytes. The laboratory is also not allowed to refuse samples based on matrix, unless the matrix is outside the matrices defined in their awarded Analyte Groups. If the laboratory determines that the samples would contaminate their laboratory or analytical systems, such samples can be subcontracted to a third-party laboratory. The cost for said analyses will be the line item price previously specified in the contract. No up-charge for subcontracting shall apply.

3. If subcontracting is deemed necessary, the Department must be notified, within 24 hours of the receipt of the sample, and provided with the name of the Subcontractor and a complete list of the tests to be performed by the Subcontractor. Once notified, the Department will approve or disapprove of the proposed subcontracting proposal. The designated Subcontractor's laboratory must meet all requirements, including insurance requirements, applicable to the primary contractor. All technical requirements of the contract and the PAP with regard to the performance of the analysis will remain in effect for the subcontract laboratory. This includes, but is not limited to, holding times, QA/QC, and reporting deadlines. The Department may require a performance evaluation and may disapprove the use of a specific Subcontractor. All analytical results generated by a Subcontractor must be reported to the Department by the primary Contractor, in the format required by the PAP. Under no circumstances should the primary Contractor report the work of the Subcontractor as their own. Results derived from the Subcontractor must be clearly identified as such.

A-10. Data Confidentiality: All data submitted and generated under Analyte Group 2 contracts must be treated as confidential. Any information released to the contract laboratory by the Department regarding sample origin and collection cannot be shared with outside parties, including subcontracted laboratories. Additionally, all data generated by the laboratory as a result of sample processing and analysis cannot be released to outside parties. Under no circumstances should data submitted to the laboratory by the Department, or data generated by the laboratory on the behalf of the Department be disseminated to outside parties without the written consent of the Department.

A-11. Record Retention: All records submitted or generated under the Analyte Group 2 contracts must be retained for a minimum of five (5) years from the date an acceptable data package was delivered to the Department. All information necessary for the historical reconstruction of data must be maintained by the laboratory for the five-year period. Records that are stored only on electronic media must be supported by the hardware and software necessary for their retrieval and reconstruction. This is a minimum retention period; contractors are encouraged to maintain archives on a longer term when practical and feasible.

## EXHIBIT B – Data Delivery Requirements

B-1. General Reporting Specifications:

A. Unless otherwise specified, and arranged in writing by the NYSDEC Project Manager, all samples for the purpose of this Analytical Group may be used in regulatory decisions and data reports are to be complete and legally quantitative. Raw data, chromatograms, and bench logs are to be retained for a minimum of 5 years (See Exhibit A-11).

B. Unless stated otherwise, send one copy of all data and reports, in electronic format to the NYSDEC Analyte Group 2 Contract Manager and the designated Project Manager as specified on the chain of custody and cover letter accompanying the samples.

C. The data package reported by the laboratory is to be delivered in portable document format (PDF, password protected against modification, but printable) with the electronic data deliverable (EDD; see details in Exhibit B-3). The data package and EDD should be transmitted by e-mail to the Project Manager or made available for download on a secure site operated by the laboratory.

D. Provide the standard operating procedures used for the analyses via e-mail concurrently with the first analysis report prepared under this contract, or when requested by DEC staff. Include calculation methods for determining analyte concentrations where relevant. Following this initial submission, provide the SOPs on March 31 of each year in which analyses were performed during the previous 12 months.

E. Detection and Reporting Limit Definitions and Requirements:

The NYSDEC offers these definitions and explanations of terms for Analyte Group 2:

1. The minimum detection limit (MDL) is defined as in 40 CFR Part 136 Appendix B, Revision 2: the minimum measured concentration of a substance that can be reported with 99% confidence that the measured concentration is distinguishable from method blank results. As stated in the IFB (Article 4(a)), vendors must include copies of MDL study results for all target analytes in a tissue or simulated tissue matrix. These studies must include information on calibration data, instrumental conditions, method blanks, and QA/QC, with upper and lower control (acceptance) limits. Any updates to the MDL will be reported to the NYSDEC Contract Manager.
2. The sample detection limit (SDL) is defined as the MDL, adjusted for sample-specific actions, such as dilution, aliquot size, matrix effect, or recovery. It should be determined for every analysis run. This is the preferred lower limit of detection level. In addition to the MDL study, contractors must submit an explanation of how the SDL is determined.
3. The lower limit of quantification (LLOQ), limit of quantification (LOQ), reporting limit (RL), estimated quantification limit (EQL), and practical quantification limit (PQL) are largely synonymous and defined as the lowest level that can be reliably achieved within the specified limits of precision and accuracy during routine laboratory operating conditions (50 FR 46902; U.S. EPA [2014a] SW-846). Method

8000D (U.S. EPA 2014b) in SW-846 can be used to determine and verify the LLOQ. The U.S. EPA-RCRA program will be recommending using the LLOQ for reporting in future updates to SW-846 (80 FR 156: 48522-48528). The NELAC Institute (TNI) LOQ standard is also acceptable (Volume 1, Module 4, 1.5.1 and 1.5.2). Vendors should include copies of study results used to determine and verify the LLOQ. Verification of the LLOQ should include at least seven (7) replicates at the LLOQ. Whichever method is used, it must be clearly defined and documented.

4. The sample quantification limit (SQL) is defined as the LLOQ, adjusted for sample specific actions, such as dilution, aliquot size, matrix effect, or recovery. It should be determined for every analysis run. This is the preferred lower limit of quantification reporting level. In addition to the LLOQ verification, vendors must submit an explanation of how the SQL is determined.
5. The contract required quantification limit (CRQL) is defined as the target minimum level of required quantification acceptable under the IFB and Price Tables (Appendix 1).
6. The LLOQ must be less than or equal to the CRQL.
7. Results above the SDL, but below the SQL, must be flagged/qualified as estimated ("J"). Results below the SDL should be flagged/qualified as not detected ("U"). The laboratory must indicate whether they are reporting the statistically derived MDL or the instrument run and matrix derived SDL. These qualifiers must not have any other uses or meanings without additional specifically defined modifiers.

B-2. Data Package Requirements: The following elements are to be included in a single Adobe Portable Document Format (PDF) file. The PDF file is to be password protected from alteration but should allow text to be copied and allow printing. Any replacement files needed due to corrections must have names that clearly indicate the version using, for example, a date or a version suffix. Report elements are desired to appear in the file in the order listed below and must be bookmarked. Searchable text rather than images is preferable when reporting computer generated output.

A. Cover page:

1. Laboratory name.
2. NYSDEC contract number.
3. Laboratory project or case name.
4. Sample delivery group (SDG) number.
5. NYSDEC project name.
6. NYSDEC project manager (NYSDEC person to whom report is delivered).
7. Date of preparation of sample data package.
8. List of NYSDEC sample ID numbers included in each SDG in alphanumeric sort order, including a cross-reference to laboratory sample IDs; use a separate page if appropriate.

B. Case narrative:

1. General summary: Describe the analysis in summary form including any problems observed, sample condition upon receipt, analytical concerns, and quality control

issues. Describe the condition in which samples were received at the laboratory including thermal status (frozen, partially thawed, etc.), noting and discussing any problems. Document the holding time for samples contained in the SDG.

2. Methods: List sample preparation and analytical methods. Include the method accuracy and precision for each target analyte.
3. Preparation: State whether sample preparation proceeded normally; describe any exceptions or problems encountered. The discussion should include all phases of sample handling from processing the tissue to final analysis.
4. Quality control: Describe and discuss any failures to meet laboratory or contract criteria or any other anomalies.
5. Reanalysis: State whether any reanalysis was required for any samples in the SDG and explain why any reanalysis was needed. If reanalysis was required, list the NYSDEC sample ID and laboratory sample ID numbers of reanalyzed samples. For each reanalyzed sample, provide all results, including QC information, and state the analysis in which the laboratory has the highest confidence.
6. Gas chromatography columns: Provide gas chromatography (GC) column identification including the brand name, internal diameter (in millimeters), length (in meters), and any additional pertinent information about the column.
7. Mass spectrometer: Provide a description of the mass spectrometer used in the analysis, including brand, model, system specification, and operating conditions.
8. Comments (optional): Include any other information useful in interpreting the report and data. Any relevant changes in procedures or equipment from previous results reported under the contract should be described.
9. Certification: Conclude the SDG narrative with the following statement, verbatim: "I certify that this data package is in compliance with the terms and conditions of the contract, both technically and for completeness, for other than the conditions detailed above. Release of the data contained in this Sample Data Package and in the electronic data deliverables has been authorized by the Laboratory Manager or his/her designee, as verified by the following signature." This statement is to be directly followed by the signature of the Laboratory Manager or designee with a typed line below it containing the signer's name and title, and the date of signature. This electronic signature will be considered a legally binding signature under New York State's Electronic Signatures and Records Act.

C. Sample summary page

1. Header information: Laboratory name, NYSDEC project name, and any other identifying information that the laboratory wishes to include.
2. Sample information, one row for each sample: Required – NYSDEC sample ID number, laboratory sample ID number. Optional: fish species name or code, date of sample collection, analyses performed.

D. Chain of custody form with all signatures.

E. Sample receipt form: If not indicated on the chain of custody forms, provide a copy of the forms or records used to log and document samples upon receipt.

F. List of codes and abbreviations: List and define all sample qualifier codes and other abbreviations used in the report. Include any laboratory codes, such as qualifiers, used in the electronic data deliverable.

G. Correspondence: Include copies of relevant correspondence concerning procedures, problems, decisions, or other information needed to document the analytical process.

H. Analytical results: For PCB results, provide a summary for each sample with all analytical results and ancillary information such as extraction date, analysis date and sample mass. Provide separate sections for percent lipid and percent moisture with processing information for the samples. Percent lipid and percent moisture results can be presented either in a summary table with all results or along with the PCB results for each sample. Include the SDL and SQL for each analyte. If any sample dilutions are required, report each analyte using the smallest dilution for which the results are acceptable.

I. Quality control:

1. Summarize results from method blank, surrogate, matrix spike and duplicate analyses.
2. Provide detailed reporting on quality control analyses. Report duplicate sample results along with relative percent difference for any duplicate analyses. Report method blank results. Report matrix spike amount, total recovery, net recovery and percent recovery. For reference material analyses, report the reference values if known, results of the reference sample analysis, and the percent recovery of the analyzed reference sample.
3. Provide a table with summary surrogate recovery information for each sample. Include the acceptable range of surrogate recoveries.
4. If any sample dilutions are required, this must be indicated in the narrative. The vendor must also state which results they have the most confidence in and are thus reported.

J. Raw data: For each sample, including quality assurance samples, provide the raw data including the chromatograms, the instrument analytical report and the integration table. Provide an example calculation.

K. Calibration data: Provide instrument calibration data including chromatograms and all associated quality assurance data. This should include the most recent initial calibration completed before sample analysis as well as the continuous calibration applicable to the analyzed samples. Calibration plots are desirable but not required.

L. Run tables and logs: Provide sample run tables for calibration and analysis. Provide extraction logs. Provide information on standard batches and sources.

B-3. Electronic Data Deliverable:

A. Acceptable file formats for electronic data deliverables are comma separated value (CSV, preferred), Microsoft Access, Microsoft Excel, and other spreadsheet formats readable by

Microsoft Excel. CSV and spreadsheet files must contain column headers. Files submitted in CSV form must be readable by Microsoft Excel and must preserve all formatting traits (column headers, data position, etc.) when viewed with the Excel software. By mutual agreement between the reporting laboratory and the NYSDEC project manager, an XML file format or the NYSDEC EQuIS data format may be used. The same file naming convention is to be used to name all files sent by the laboratory. File names may not contain spaces. Any replacement files needed due to corrections must have names that clearly indicate the version using, for example, a date or a version suffix.

B. Data are to be in long format with a single record for each analytical result. Data, including quality assurance results, are to be reported in a single file as specified below. The following data values are to be reported in the order in which they are listed below. Each entry below has the field name, with the desired the field type and recommended length in parentheses (e.g., "Text 20" for a text field of 20 characters), the definition of the field, and required values, if any.

1. TAGNO (Text 20): DEC sample identification number; note that this is a character field and that leading zeros in the sample number must be included.
2. LABNO (Text 20): Laboratory sample identification number.
3. SPP (Text 7): Species code or name as supplied by DEC.
4. Sex (Text 1): Fish sex if determined by the laboratory (M = male, F = female, U = not determined).
5. Weight (Double 8): Weight in grams of the fish or sample as received.
6. Analysis_Date (Number 8): Date of analysis formatted as YYYYMMDD.
7. Analysis_Time (Number 4): Time of analysis formatted as HHMM, based on a 24-hour clock.
8. Sample_Type (Text 15): Distinguish submitted tissue samples, reference material, blanks, duplicates, etc., using appropriate codes
9. Prep (Text 3): For fish samples received whole at the laboratory, the code for method of preparation as specified on the sample transmittal documents (HV = head and viscera removed, SF = standard fillet, WH = whole organism). For fish samples received as a fillet, use code SF for standard fillet. For fish samples received as homogenate, use code HOM for homogenized. Leave null for laboratory samples. Consult with the NYSDEC project manager for the appropriate code for other tissues or preparations.
10. Analyte (Text 20): Name of analyte or parameter reported on.
11. Result (Double 8): Analytical result (must be numeric). Wet weight basis.
12. Detect (Text 1): Y if detected, N if not detected.
13. Result_Qualifer (Text 5): Laboratory qualifier if any (B = blank qualified, J = between SDL/MDL and SQL, U = below the SDL/MDL, other values as defined in the laboratory report, null if no qualifier).
14. Result_Unit (Text 10): Unit of measure used for result and detection limit (metric values such as mg/kg; not ppm).
15. Sample_Detection_Limit (Double 8): Lowest concentration at which the laboratory can detect the presence of the analyte in the sample, even if not quantifiable (same units of measure as Result). The SDL is defined in Exhibit B-1.E.2.

16. Sample_Quantification_Limit (Double 8): Laboratory limit of accurate quantification (same units of measure as Result). The SQL is defined in Exhibit B-1.E.4.
17. SDG (Text 20): Sample delivery group of 20 or fewer samples associated with quality assurance analyses. This field must support an association between quality assurance work and a specific set of tissue samples.
18. Remarks (Text 50): Any information, including interpretive comments, the laboratory wants to provide (may be null).
19. CAS_Number (Text 12): CAS Registry Number if available.

C. The laboratory may include additional data fields after the required data fields.

D. Data are to be sorted using the NYSDEC sample identification number (TAGNO) as the primary sort key and the analyte (Analyte) as the secondary sort keys.

E. Results of duplicate sample analyses should be included and clearly labeled using, for example, the laboratory sample ID.

F. Include results from method blank (MB), laboratory control sample/laboratory control sample duplicate (LCS/LCSD), matrix spike/matrix spike duplicate (MS/MSD), and reference material analyses (SRM or CRM). All such quality control analyses should have a unique laboratory sample ID. The sample delivery group with which the quality control analyses are associated is to be indicated in the SDG field.

G. All numbers are to be reported with the appropriate number of significant digits. Spreadsheet formatting cannot be used to control the display of significant digits.

## EXHIBIT C – Contract Required Quantification Limits (CRQL)

C-1. General:

A. The values in these tables are target quantification (i.e., reporting) limits, not absolute detection limits. It is expected that laboratories will strive for, and regularly attain, better performance (i.e., lower reliable reporting limits). Specific quantification limits are highly matrix dependent. It is expected that the laboratory will make every effort possible to meet the quantification limits listed herein but it is realized that these limits may not be achievable in all instances.

B. Results for tissue samples should be reported on a wet weight basis.

C. Percent moisture and lipid must be determined on a balance that reads to the nearest 0.0001 g, with a quantification limit of 0.01%.

D. Changes to the Target Compound Lists (TCLs) (e.g., adding an additional analyte) or lower CRQLs may be requested under the flexibility clause in the contract.

C-2. The Contract Required Quantification Limit (CRQL) for the target analytes in Analyte Group 2:

A. PCB congeners by GC/LRMS methods: Actual quantification values achieved depend on the method employed. An examination of PCB congener analyses that used low resolution methods (e.g., Coots 2014; Van Bavel et al. 1995) and previous data delivered to NYSDEC suggest that appropriate CRQL values should be less than 1 ng/g for each individual congener or group of coelutant congeners. Laboratories must report all quantified congeners, as well as homolog groups and a total PCB value.

B. Coplanar/non-*ortho* and mono-*ortho* substituted PCB congeners by HRGC/HRMS: The CRQLs for individual PCB congeners are the Minimum Levels of Quantification (MLs) for solids in ng/kg (ppt) as listed in Table 2 of U.S. EPA (2010).

C-3. Procedure Labs Must Follow If and When a Specified CRQL Cannot Be Met: If a CRQL cannot be met, analysis of samples should be suspended. The laboratory should follow the corrective actions specified in the approved method or SOP and must notify the NYSDEC Project Manager within one (1) week if corrective action is not successful. Any deviation from the approved method or SOP must be approved by the NYSDEC Project Manager.

## EXHIBIT D – Methods

D-1. General: A performance-based analytical program is recommended for the analysis of target analytes. Unless a specific method of analysis is stipulated in the Analytical Group Price Table, bidding laboratories should propose methods for detecting and measuring the target analytes in biological tissue samples at the time of response to the IFB. Standard methods approved by EPA, FDA, ASTM, etc., should be used whenever possible. If methods other than standard methods are anticipated or proposed, including variations to standard methods, such alternatives must be submitted to the Department for review and approval at the time of response to the IFB and must be accompanied by proof of method performance. The recommended method will be acceptable if it can be demonstrated to produce an analytical result that is consistent with quality control requirements outlined in Exhibit E and the CRQLs described in Exhibit C. Once the contract has been let, approved SOPs will be appended to this Prescribed Analytical Procedure (PAP). This recommendation is based on the assumption that the analytical results produced by different laboratories and/or different methods will be comparable if appropriate quality control procedures are implemented. This approach is intended to encourage the use of new or improved analytical methods without compromising data quality (U.S. EPA 2000).

D-2. Standard Methods: Following are a summary of standard methods known to be acceptable for detecting and measuring the analytes in this Analyte Group. Multiple extraction and cleanup methods can be used in conjunction with these instrumental methods to achieve the performance standards required for this Analyte Group:

A. PCB congeners by GC/LRMS: The low-resolution variety of MS analysis was the basis of the EPA Method 680 (U.S. EPA 1985, Beliveau 2001). Modified Method 8270D (U.S. EPA 2014c) has also been used as the basis for LRMS PCB congener analyses. These methods are described as partial congener analysis (Beliveau 2001; Coots 2014). Full congener analysis is nearly impossible without having coelution of some peaks (Beliveau 2001).

B. Method 1668C (U.S. EPA 2010) for the analysis of chlorinated biphenyl congeners in water, soil, sediment, biosolids, and tissue by HRGC/HRMS.

D-3. PCB Analytes of Interest: There are 209 possible PCB congeners. The desired result is quantification of the highest number of congeners, either as individual congeners or coelutants. Twelve (12) of these are designated as “dioxin-like” highly toxic congeners (U.S. EPA 2010). Because of the high toxicity and concern about these WHO-listed congeners, NYSDEC is requesting further analysis and more detailed quantification of the coplanar/non-*ortho* substituted (PCBs 77, 81, 126 & 169) and mono-*ortho* substituted (PCBs 105, 114, 118, 123, 156, 157, 167, & 189) PCB compounds in this Analyte Group. Because of the possible future inclusion in the WHO TEF scheme, quantification of PCB 37 is also desirable (Van den Berg, et al. 2006). A performance-based approach is required for PCB congener analysis.

## EXHIBIT E – Quality Assurance / Quality Control

E-1. In order to meet the Department goal of 100% usable and defensible data, a robust quality assurance/quality control plan is required. This includes laboratory controls for accuracy (results represent the true value for a sample), precision (results are consistent), and completeness (results are usable for each project). Contracted laboratories are required to develop a Quality Assurance Management Plan, analyze quality control samples, and meet initial and ongoing acceptance criteria. Provided there is adequate sample material, any SDGs failing QC controls will be re-run at no cost to the Department. All data delivered to the Department will be reviewed for usability.

E-2. Quality Control Acceptance Criteria for Analyte Group 2: Acceptance criteria for individual QA/QC samples are listed in Table 6 of U.S. EPA. (2010) for PCB congeners. Limits for estimated precision as relative percent difference (RPD) between duplicate samples is ≤ 30% (≤ 50% if a target is less than 5 × MDL). Limits for lipids and moisture estimated accuracy as percent recovery are ± 20%, and estimated precision as RPD in duplicates is ≤ 20%.

E-3. Other methods and analytes have similar guidance for acceptable ranges of recovery on reference material and spikes, and relative percent difference on duplicates. Contractors will state what ranges they are using in their SOPs and cite justification. It is important to note that these are the acceptable values for individual quality control samples. As part of the required Quality Assurance Management Plan (below, Exhibit E-5), contractors will propose their ongoing performance control tracking measures, including determination of warning and control limits, work stoppage rules, and corrective procedures.

E-4. As evidence of acceptable overall accuracy, in addition to meeting the above requirements for individual quality control samples, matrix spike recoveries and lab control sample recoveries must average between 85% – 115%, as demonstrated by a minimum of ten analyses conducted as part of the contractors' ongoing quality assurance program. The contractor must also achieve average percent recoveries on certified reference material that are within ±20% of the certified value..

E-5. Quality Assurance Management Plan (QAMP): At the time bids are submitted, bidding laboratories must submit a Quality Assurance Management Plan that describes the general and specific procedures used within the laboratory to achieve scientifically valid and legally defensible data. The QAMP should describe the policies, organization, objectives, functional guidelines, and specific QA and QC activities of the laboratory, designed to achieve the data quality requirements when running performance-based methods. The QAMP should include, as a minimum, the following elements:

A. QA policy, objectives, commitments, and any allowable departures from documented policies.

B. Organizational structure and personnel, including descriptions of key personnel; identify the relationship between management, operations, support, and QA personnel.

C. A description of the facility and equipment used.

D. A description of document control procedures, such as sample tracking and custody procedures, and a general description of logs and records maintained in conjunction with sample analyses.

E. Description of relevant QC procedures, such as methods implementation, calibration procedures and frequency, standards preparation procedures, traceability of measurements, decision processes, internal QC checks and procedures, corrective action implementation, and verification of electronic data management systems.

F. Data validation procedures.

G. Determination and monitoring of method QA procedures.

E-6. General Procedures for Quality Control:

A. Standard QA/QC methods and procedures specified in Section 8 of U.S. EPA (2000) and Section 9 in U.S. EPA (2010) must be complied with, as appropriate. Standards and surrogates must be obtained from recognized commercial sources and meet the quality and purity requirements described in U.S. EPA (2000).

B. An analytical batch is defined as one Sample Delivery Group (SDG). Each SDG will contain 20 field samples, except for the final SDG that might contain fewer, if the total number of individual field samples submitted in a delivery was not evenly divisible by 20.

C. The laboratory must implement procedures for documenting the effect of the matrix on method performance (precision, accuracy, method sensitivity). At a minimum, this should include the analysis of QC samples including a method blank, a matrix spike when applicable to the method, a duplicate (either a field sample duplicate or matrix spike duplicate), and a laboratory control sample (LCS) in each analytical batch and the addition of surrogates to each field sample and QC sample when surrogates are used. Any method blanks, matrix spike samples, and replicate samples should be subjected to the same analytical procedures as those used on actual samples (U.S. EPA 2000).

D. Matrix Spikes (MS), Matrix Spike Duplicates (MSD): In accordance with (U.S. EPA 2000), at least one MS and MSD should be run with each analytical batch. If samples are expected to contain target analytes, then laboratories should use one matrix spike and a duplicate analysis of an unspiked field sample. If samples are not expected to contain target analytes, the laboratories should use a matrix spike and matrix spike duplicate pair, spiked with the appropriate standard(s). DEC's default is a duplicate, but the DEC project manager will determine if an MSD or un-spiked field sample duplicate is to be run. The MS/MSD concentrations should be distributed uniformly throughout the calibration range of the instrument.

E. If isotope dilution methods are used for recovery estimation and correction, MS/MSD samples are not required. These should be replaced with a Field Duplicate – re-analysis of a separate aliquot of tissue sample. In this case field duplicates will be run at a 5% rate (one per 20 samples).

F. An extract of the ongoing precision and recovery (OPR) aliquot must be analyzed before the samples from the same batch. For PCB congeners, Table 6 of U.S. EPA (2010) provides acceptance ranges as percent recoveries for OPR aliquots, and acceptance ranges as percent recoveries for IPR aliquots. If all compounds in the OPR extract meet acceptance criteria analysis of blanks and samples may proceed.

G. Method blanks are required to demonstrate freedom from contamination (U.S. EPA 2014b). A method blank must be analyzed at least once for every 20 samples (5%). Method blanks must be subjected to the same analytical treatment and procedures as field samples. Matrix matching of the blank is not allowed. The method blank should be analyzed immediately after the OPR aliquot to confirm lack of carryover from the OPR analysis.

1. The contractor will not subtract the results of the blank from any associated samples.
2. Blanks are considered acceptable if target analyte concentrations are less than the MDL; however, blanks may be considered acceptable if concentrations are below the LLOQ or all samples in the SDG contain concentrations that are ≥10 times the blank concentration for all analytes.
3. Regular blank concentrations above the MDL, even in cases where high sample concentration allows acceptance, constitute a laboratory contamination issue and corrective action must be taken by the contractor.
4. Method blanks for percent lipid must be <0.03%.
5. If method blank results are not acceptable, the vendor will take corrective action to locate the source of the contamination. Affected samples must be re-extracted and re-analyzed. If re-analysis is not practical, qualification of then sample data will be qualified as “B”, probable blank contamination.

H. The laboratory shall spike all samples with labeled compounds to monitor method performance. The recovery of each labeled compound must be within the limits specified in Table 6 of U.S. EPA (2010) for PCB congeners.

I. As stated in Exhibit B-2.I.4, if dilution of a sample (field or Matrix QC) is required, results from the instrument run(s), including surrogate recovery, and any notes used in determining the need and rate of dilution will be included in the data package. The contractor shall state which dilution, and which analytes in each, they have the most confidence in.

J. Calibration:

1. Initial calibration or linear calibration must be performed at least once per year (or at the minimum frequency prescribed in the proposed method if that is more frequent). This frequency will be defined in the QAMP. The concentrations at which a

laboratory is able to detect and measure analytes are determined during the initial calibration of the analytical instrument, using either internal or external calibration standards. As long as the instrument's operating condition remains constant and the instrument remains free of contaminants, the identification and quantification of analytes should continue to meet or be lower than CRQLs.

2. Continuing calibration verification (VER) must be verified at least each shift by analyzing a verification standard. Corrective action must be taken according to the method or vendor SOP if the VER fails to meet method or SOP requirements. If over 10% of the analytes in a multi-analyte method exceed the VER criteria, a new calibration is required (U.S. EPA 2014b).
3. Additionally, in accordance with U.S. EPA (2000), a VER standard should be included after 10 – 12 field samples in the analysis sequence as a calibration check. Injections of method blank extracts, matrix spike samples, and other non-standards are counted in the total. Solvent blanks, injected as a check on cross-contamination, need not be counted in the total. The response of calibration verification standards shall be monitored in control charts for signs of deterioration so that corrective action can be taken as soon as possible. The response factors for the calibration must be within ±20% of the initial calibration (U.S. EPA 2014b).
4. The concentration of the verification standard (VER) is equivalent to the mid-range concentration (CS3) of the five calibration standards.
5. The m/z abundance ratios must be within the range specified in Table 8 of U.S. EPA (2010) for PCB congeners.
6. If the VER does not meet the ±20% limit, the laboratory will stop analyses and take corrective action. If the response for the standard is still not within ±20%, then a new initial calibration must be performed (U.S. EPA 2014b). Once recalibrated so that performance standards are met, samples that were analyzed when the calibration was questionable should be reanalyzed. If it is not possible to reanalyze the samples, those data will be flagged. If the new calibration results in detection/quantification values for analytes that do not meet CRQLs, then the equipment shall be taken off-line for maintenance and/or decontamination. See section 11.11 of U.S. EPA (2014b) for suggested chromatographic system maintenance guidance.

K. Part of a robust quality control process is consistent tracking of QC and ongoing performance review results. In addition to the performance control tracking submitted with the bid proposal (IFB Article 4(a)), NYSDEC may request a quality tracking report which includes copies of control charts used to assess analytical performance. This will include items such as Shewart charts, I-MR charts, and Xbar-R charts (U.S. EPA 2000; Mullins 2003; Konieczka and Namieśnik 2009) and a narrative for matrix spike recoveries, duplicate percent differences, blank results, and reference material recoveries for all instruments used in the analysis of NYSDEC samples. This should also include information in order to group data by the analyst/technician operating the instrumentation. Determination of Center Line (expected values), upper and lower warning limits (UWL, LWL) and upper and lower control levels (UCL, LCL) must be documented. Control charts and narrative should be provided to the NYSDEC Contract Manager electronically in PDF and spreadsheet formats.

E-7. A QC Check Sample should be analyzed at least quarterly from a source independent of the calibration standards. Ideally, the QC Check Sample should be a certified reference material containing the target analytes in known concentrations in a matrix similar to fish tissue. Certified tissues are available from multiple national and international agencies (e.g., NIST, NRC Canada, IRRM, etc. Also see Ricci et al., 2016). If a suitable reference material is not available the vendor must contact the NYSDEC Project Manager for approval of an alternative. Results from QC Check Samples will be supplied to NYSDEC as per Exhibit E-3.K. QC Check Samples are not billable.

E-8. Standard Reference Material (SRM): Reference material will be run at a rate of 1 sample for every 20 field samples submitted for analysis of PCBs. DEC may provide Hudson Reference Material (HRM) or other suitable reference material (e.g., NIST 1947) may also be provided for a final rate of 1 reference sample per 20 field samples (5%; U.S. EPA 2000). Analysis of NYSDEC supplied SRM samples are billable.

E-9. The Department may, on occasion, submit QC samples that are blind to the laboratory. These may be split samples also analyzed at other laboratories, duplicate samples, spiked samples, or certified reference material. These will be billable samples, and the laboratory will be notified of their performance after analysis is complete.

E-10. MS, MSD, SRM, and duplicate analysis of field samples are billable. Method blanks and LCSs and other internal laboratory quality control analyses are not.

## EXHIBIT F – Sample Preparation, Shipping, Handling, and Storage

F-1. Sample collection, initial storage, and shipping will be the responsibility of the NYSDEC. Samples will be primarily fish tissues. However, other biological tissues may be submitted, and the processes will be equivalent. Sample chain of custody shall be maintained and recorded on a signed chain of custody form. Samples will be individually weighed, measured, tagged, and sealed in a plastic bag. The plastic bag will be marked with a manila tag containing capture data (project, date, location) and the sample tag (ID) number. Sample data will be recorded on a field collection log and chain of custody form.

F-2. Sample Preparation: Tissue samples will be prepared for analysis in accordance with NYSDEC Prep Lab 4.1 Standard Operating Procedure (attached as Appendix A: SOP PREPLAB4.1.doc dated 3/09/2016), which is supplemented but not superseded by section 7.2.2 of U.S. EPA (2000). Fish may be delivered to the vendor either as whole fish, fillets, or homogenate. If delivered as whole fish, the vendor is responsible for fillet and homogenate preparation. The service is billable as defined in the IFB Group Price Tables. Homogenization of fillets or whole fish is included in the analysis price as specified on the cost Price Tables. Sex determination and assessment for morphological abnormalities (sections 7.2.2.3 – 7.2.2.5 of U.S. EPA (2000) are not required but may be requested by NYSDEC at the time of shipment.

F-3. Sample Receipt Verification Report:

A. All samples are sent on dry or water ice to contract analytical laboratories using overnight delivery. The receiving contract laboratory is notified either by telephone or by e-mail message upon shipment and is required to confirm receipt and condition of the samples upon arrival.

B. Within one week of sample receipt, the laboratory is required to send a Sample Receipt Verification Report that includes the laboratory's signed chain of custody forms with sample identification data, date received, matrix, and analyses. The sample status report must also show:

1. Date, DEC project name, laboratory assigned SDG, sample submitter, matrix, estimated price and analysis information.
2. Sample ID numbers matching NYSDEC collection records and chain of custody forms.
3. All samples intended for analysis are included.
4. Samples are grouped in batches of 20 for quality assurance purposes with batches uniquely labeled.
5. Requested analyses.
6. Requested sample preparations.

C. Sample processing cannot begin until the NYSDEC Project Manager acknowledges receipt of the Sample Receipt Verification Report and confirms its correctness.

F-4. Discrepancy Reports: If any problems are noted by the receiving contract laboratory upon receipt of the samples, the contract laboratory must contact the NYSDEC Project Manager by telephone or by e-mail. Examples of discrepancies that would require immediate resolution include:

A. If sample material was not frozen upon receipt, discuss its condition with the NYSDEC Project Manager and determine usability. Samples that are still cold (solid) and that have not leaked fluids will generally be analyzable while samples that have lost fluids or warmed will be discarded. The condition and final disposition of unfrozen samples should be reported in writing within one week of receipt.

B. A discrepancy in data such as the sample ID or species is found between DEC's collection records and the chain of custody sent to the laboratory.

1. Confirm that both the external tag for the sample bag and the metal identification tag, if present, on the sample were checked.
2. If the problem was other than typographical and easily fixed, summarize the resolution to the problem agreed upon with the NYSDEC Project Manager in the SDG narrative.

C. The samples received do not match those listed on the NYSDEC chain of custody form (e.g., wrong batch of samples, missing samples, extra samples).

D. Sample processing cannot begin until all discrepancy reports have been resolved and a final disposition determined between the receiving contract laboratory and the NYSDEC Project Manager.

F-5. Holding Times:

A. Samples can be held for up to one year when frozen at ≤ -20°C.

B. When samples are thawed and homogenized, they should be extracted as soon as possible. While awaiting extraction, sample material must be stored at ≤ -20°C.

C. Samples must be analyzed within 40 days of extraction. While awaiting analysis, sample material (i.e., extracts) must be stored at ≤ 6°C.

F-6. Post Analysis: Any remaining extracted sample material can be stored following injection for another 40 days at ≤ 6°C, during which time the results of the analysis are reviewed. At least 50 grams, if enough tissue remains, of homogenized biological tissue (unextracted sample material) must be stored at -20°C for two years post analytical result acceptance.

## LITERATURE CITED


Beliveau, A.F., 2001. PCB Analyses Needs for Risk Evaluations. In: Robertson, L.W. and L.G. Hansen, eds., PCBs Recent Advances in Environmental Toxicology and Health Effects. The University Press of Kentucky. Pp 424 – 428.

Coots R., 2014. PCB Method Comparison of High and Low Resolution Sediment Analysis. Washington State Department of Ecology, Toxic Studies Unit. Publication # 14-03-009. Olympia, WA.

Konieczka, P. & J. Namieśnik, 2009. Quality assurance and quality control on the analytical chemistry laboratory. CRC Press – Taylor & Francis Group, Baton Rouge, LA. 233pp.

Mullins, E., 2003. Statistics for the Quality Control Chemistry Laboratory. Royal Society of Chemistry, Cambridge, UK. 455pp.

Ricci, M., R. Lava & B. Koleva, 2016. Matrix certified reference materials for environmental monitoring under the EU Water Framework Directive: an update. Trends in Analytical Chemistry 76: 194-202.

U.S. EPA, 1984. Method 625: Base/Neutrals and Acids. Methods for Organic Chemical Analysis of Municipal and Industrial Wastewater. Appendix A to Part 136

U.S. EPA, 1985. Method 680, Determination of Pesticides and PCBs in Water and Soil/Sediment by Gas Chromatography/Mass Spectrometry, U.S. Environmental Protection Agency. On-line document: http://www.epa.gov/nscep (document #905R85112).

U.S. EPA, 2000. Guidance for Assessing Chemical Contaminant Data for use in Fish Advisories, Volume 1, Fish Sampling and Analysis, Third Edition. U.S. Environmental Protection Agency, Office of Water, EPA 823-B-00-007, November 2000.

U.S. EPA, 2007. EPA Method 8081B, Organochlorine Pesticides by Gas Chromatography Revision 1 dated February 2007, in: Test Methods for Evaluating Solid Waste, Physical/Chemical Methods. U.S. Environmental Protection Agency, EPA Publication SW-846. On-line document: http://www.epa.gov/wastes/hazard/testmethods/sw846/online/index.htm.

U.S. EPA, 2010. Method 1668C: Chlorinated Biphenyl Congeners in Water, Soil, Sediment, Biosolids, and Tissue by HRGC/HRMS. U.S. Environmental Protection Agency EPA 820-R-10-005, April 2010.

U.S. EPA, 2014a. The SW-846 Compendium, U.S. Environmental Protection Agency. On-line access: http://www.epa.gov/hw-sw846/sw-846-compendium.

U.S. EPA, 2014b. Method 8000D, Determinative Chromatographic Separations Revision 4 dated July 2014, in: Test Methods for Evaluating Solid Waste, Physical/Chemical Methods. U.S.

Environmental Protection Agency, EPA Publication SW-846. On-line document: http://www.epa.gov/wastes/hazard/testmethods/sw846/online/index.htm.

U.S. EPA, 2014c. Method 8270D, Semivolatile Organic Compounds by Gas Chromatography/Mass Spectrometry Revision 5 dated July 2014, in: Test Methods for Evaluating Solid Waste, Physical/Chemical Methods. U.S. Environmental Protection Agency, EPA Publication SW-846. On-line document: http://www.epa.gov/wastes/hazard/testmethods/sw846/online/index.htm.

Van Bavel, B., C. Naf, P-A Bergqvist, D. Broman, K. Lundgren, O. Papakosta, C. Rolff, B. Strandberg, Y. Zebühr, D. Zook & C. Rappe, 1995. Levels of PCBs in the aquatic environment of the Gulf of Bothina: benthic species and sediments. Marine Pollution Bulletin 32(2): 210-218.

Van den Berg, M., L.S. Birnbaum, M. Denison, M. De Vito, W. Farland, M Feeley, H. Fiedler, H. Hakansson, A. Hanberg, L. Haws, M. Rose, S. Safe, D. Schrenk, C. Tohyama, A. Tritscher, J. Tuomisto, M. Tysklind, N. Walker & R.E. Peterson, 2006. The 2005 World Health Organization Reevaluation of Human and Mammalian Toxic Equivalency Factors for Dioxins and Dioxin-Like Compounds. Toxicological Sciences 93(2): 223-241.

## Appendix A – NYSDEC SOP PREPLAB4.1 dated 03/09/2016

PREP LAB STANDARD OPERATING PROCEDURE
NYS DEPARTMENT OF ENVIRONMENTAL CONSERVATION

**Hale Creek Field Station**

**Name of document:** SOP PrepLab4.1 (03-09-2016)
**Revision date:** 05/28/2014
**Previous revision:** SOP PrepLab3 (03-16-2011)
**Reason for this revision:**

- File PrepLab4.1: (03-09-2016) has minor formatting changes from PrepLab4 in order to match the format of the 2016 Analyte Group 2 PAP. No functional changes were made and the file does not require a full version change. -JCB
- File Prep Lab4: Revise Section 3.B.i to indicate that the standard fillet is taken from the left side of a fish.

**Reference:** *Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories, Volume 1, 3rd edition* (U.S. EPA Office of Water, November 2000)
**Summary:** Samples are received at Hale Creek Field Station and dissected, ground and homogenized for future chemical analysis. In addition, samples for organochlorine analysis are freeze-dried to remove moisture.
**Background:**
New York State Department of Environmental Conservation conducts studies requiring chemical analysis on fish or other biological tissues. Routine monitoring and surveillance studies develop data on contaminants in fish for several reasons:

1. To identify sources of environmental contamination;
2. To identify the geographic extent of environmental contamination;
3. To identify temporal trends of contaminants in fish and wildlife;
4. To identify potential impacts to fish and their consumers; and
5. To provide information regarding human consumption advisories.

Chemical analyses of edible fish flesh have been determined to be the most appropriate analyses for satisfying all of these objectives. The following methodology has been developed in order to standardize the tissues under analysis and to adequately represent the contaminant levels of fish flesh. The portion of edible flesh analyzed will be referred to as the standard fillet unless otherwise noted. For some species, the procedure is modified as indicated below.

### 1. SAMPLE RECEIPT

A) All samples received by the lab are to be accompanied by a Collection Record and Continuity of Evidence form.
B) After comparison of samples received with the Collection Record, the Continuity of Evidence form is signed and dated.
C) The original forms are to be retained by the lab. Copies may be returned to the delivery person.
D) Depending upon sample type, the samples are to be stored locked in either the cooler or freezer.

### 2. SAMPLE LOG IN

A) All samples are assigned a unique serial Lab # which corresponds to a specific Tag # or ID # on the sample or sample container.
B) The Lab #s are to be indicated on the Continuity of Evidence form and the Collection Record.
C) From the Collection Record the Lab #, Tag #, Species, Location, Program, Length, Weight, Sex, and Age are entered into the computer Log file.

**3. SAMPLE DISSECTION**

A) Samples are removed from the freezer and allowed to partially thaw (large samples may be removed the previous night).
B) FISH: The portion of edible flesh analyzed will be referred to as the standard fillet unless otherwise noted. For some species, the procedure is modified as indicated below.
   i) Standard Fillet – Take the fillet from the left side of the fish as follows:
      a) Remove scales from the left side of the fish. Do not remove the skin.
      b) Make a cut along the ventral midline of the fish from the vent to the base of the jaw.
      c) Make a diagonal cut from the base of the cranium following just behind the gill to the ventral side just behind the pectoral fin.
      d) Remove the flesh and ribcage from the left side of the fish by cutting from the cranium along the spine and dorsal rays to the caudal fin. The ribs should remain on the fillet.
      e) Score the skin and homogenize the entire fillet.
   ii) Modifications to the Standard Fillet
      a) Four modifications of the standard fillet procedure (see b,c,d,e) are designed to account for variations in fish size or known preferred preparation methods of the fish for human consumption.
      b) Some fish are too small to fillet by the above procedure. Fish less than approximately 6 inches long and rainbow smelt are analyzed by cutting the head off from behind the pectoral fin and eviscerating the fish. Ensure that the belly flap is retained on the carcass to be analyzed.
      c) Some species are generally eaten by skinning the fish. The skin from these species is also relatively difficult to homogenize in the sample. Hence, for the following list of species, the fish is first skinned prior to homogenization:

| | |
|---|---|
| Brown Bullhead | White Catfish |
| Yellow Bullhead | Channel Catfish |
| Black Bullhead | Lake Sturgeon |
| Atlantic Sturgeon | |

      d) American eel are analyzed by removing the head, skin, and viscera; filleting is not attempted.
      e) Forage fish and young-of-year fish are analyzed whole.
C) Wildlife/Other: Generally non-fish samples that are to be prepared have already been dissected. See supervisor for appropriate instructions.
D) All dissection tools are to be rinsed, washed with soap, rinsed, rinsed with DI water and dried between each sample dissection.

## 4. HOMOGENIZATION

A) Thoroughly grind and homogenize fish fillets using a Waring commercial chopper/grinder model WCG75. Alternatively, a comparable food chopper, food processor, grinder, blender or homogenizer may be used.

B) Mix the tissue and repeat the grinding step at least two more times and until the sample appears to be homogeneous.

C) The homogenized sample is then subsampled into appropriate glass bottles. Generally 2 – 10 g is needed for metals analysis and 20 g for organochlorine analysis. For the OC sample label and weigh an empty sample bottle. Add *ca* 20 g of sample into the bottle and weigh again.

D) The bottles are capped and stored in the freezer.

E) All homogenization tools are to be rinsed, washed with soap, rinsed, rinsed with DI water and dried between each sample.

## 5. FREEZE DRYING

A) Generally samples for organochlorine analysis are freeze dried.

B) Make sure that the unit has been drained, all valves closed and the vacuum pump oil is clear and within the acceptable markings on the site vial.

C) Turn the refrigeration unit on. After the temperature OK light comes on (less than -40°C) turn the vacuum pump on. After the vacuum OK light comes on (less than 100 millitorr) the samples may be placed on the freeze dryer (make sure samples are frozen).

D) The samples are freeze dried ca 16 hours or until the samples reach a constant weight.

E) When freeze dried, the sample bottle is weighed again.

F) The sample is stored in the freezer until analysis is started.

## 6. MINIMIZING SAMPLE CONTAMINATION DURING SAMPLE PREPARATION

A) Conduct all work in a clean environment, preferably a laboratory setting. All work surfaces, utensils and grinder work bowls and covers should be cleaned with soap and water, then rinsed with clean water, prior to working with samples, between each sample, and upon completion of sample preparation for the day. Alternatively, between samples aluminum foil may be placed on the work surface for the succeeding fish sample; discard foil after one use. DO NOT use aluminum foil if metals analyses are to be conducted on the sample.

B) Wear a clean laboratory coat for protection of clothing. Wear nitrile or latex gloves at all times while preparing samples. Clean gloves with soap and water between each sample, or discard gloves between samples and place new gloves on hands. If a glove is torn or punctured, immediately discard the glove and replace with a new glove. Discard gloves at the end of the day, or earlier if they become unsuitable for clean preparation of samples.

C) Rinse fish or other biological samples in clean water if soil, debris or other matter are evident on the exterior surfaces. Allow water to run off and dry exterior surface.

D) Following preparation of sample portions, place sample in clean containers of suitable size for the sample. For example, place small samples in chemically clean

glass jars, cover and label immediately. Jars should have PTFE-lined caps and be pre-cleaned and certified to meet EPA standards for metals, pesticides and semi-volatiles. For large samples (e.g., a fish fillet), wrap in hexane-rinsed aluminum foil and label externally. Place foil wrapped sample in a labeled food-grade plastic bag for subsequent storage and transport. If hexane-rinsed aluminum foil is unavailable, and samples are not to be analyzed for phthalates, the excised sample may be placed in a food grade plastic bag, labeled externally and placed in frozen storage. DO NOT use aluminum foil if metals analyses are to be conducted on the sample.

SOP PREPLAB4 (05-28-2014).doc
05/28/2014
Anthony J. Gudlewski