Standard Operation Procedure for:

ANALYSIS OF ORGANOCHLORINE PESTICIDES AND POLYCHLORINATED BIPHENYLS (PCBs) IN FISH TISSUE BY GAS CHROMATOGRAPHY WITH ELECTRON CAPTURE DETECTION (GC-ECD)

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1. SCOPE AND APPLICATION

1.1. This method is applicable to the determination of up to 26 organochlorine pesticides and 19 PCB congeners in fish tissue as listed in Table 3. The interferences caused by the organic matter in fish tissue require the use of appropriate cleanup procedures (see sample extraction SOP GC-004-1 Ref 15.1). The Method Detection Limits and Practical Quantitation Limits yielded by this method are provided in Table 3.

1.2. This method is based on EPA methods 8081B (Ref 15.2), 8082A (Ref 15.3 and Ref. 15.4.)

1.3. DEP Laboratory Information Management System (LIMS) test: T-PCB and T-PEST-CL, T-PCBCL-A

1.4. This Standard Operation Procedure is restricted to an analyst experienced in the use of a gas chromatograph, the interpretation of gas chromatograms, and the use of appropriate computer programs. The minimum qualifications required are:

- B.S. in Chemistry or Biochemistry with minimum one year of professional chemical or biochemical experience
- Knowledge of principles and practices of Gas Chromatography
- Ability to perform basic laboratory mathematics
- Knowledge of EZChrom Elite Client/Server software, Target chromatography software, and FL DEP LIMS.

2. SUMMARY OF THE METHOD

2.1. Fish samples are extracted using the appropriate procedure mentioned in Section 8 of this SOP. The extraction step is followed by at least one of the clean-up procedures for removal of fat tissue and other organic tissue products. The method using H_2SO_4 clean-up was validated for resistant chlorinated pesticides and 19 PCB congeners (see Table 3). The method using GPC/Florisil was validated for all chlorinated pesticides and 19 PCB congeners (see Table 3). . For acid resistant analytes the H_2SO_4 clean-up is considered superior. Therefore only non-resistant analytes are routinely analyzed using GPC/Florisil clean-up samples. The extracts are analyzed using a Gas Chromatograph (GC) equipped with two capillary GC columns and two Electron Capture Detectors (ECDs) to provide confirmatory analyses for all positive hits.

3. INTERFERENCES

3.1. Method interferences may be caused by contaminants in solvents, reagents, glassware, and other sample processing hardware that lead to discrete artifacts and/or elevated baselines in gas chromatograms. All of these materials must routinely demonstrate to be free from interferences under the conditions of the analysis by running laboratory reagent blanks as described in Section 9. Glassware must be scrupulously cleaned. Clean all glassware as soon as possible after use by rinsing with the last solvent used in it. Solvent rinsing should be followed by detergent washing with hot water, and rinses with tap water and distilled water. The glassware should then be drained dry, and heated in a muffle furnace at 400 °C for several hours. Volumetric glassware should not be heated in a muffle furnace. After drying and cooling, glassware should be sealed and stored in a clean environment to prevent any accumulation of dust or other contaminants. Store inverted or capped with aluminum foil. The use of high purity reagents and solvents helps to minimize interference problems. Purification of solvents by distillation in all-glass systems may be required. Interferences by

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phthalate esters can pose a major problem in pesticide analysis when using the electron capture detector. These compounds generally appear in the chromatogram as large late eluting peaks. Common flexible plastics contain varying amounts of phthalates. These phthalates are easily extracted or leached from such materials during laboratory operations. Cross contamination of clean glassware routinely occurs when plastics are handled during extraction steps, especially when solvent-wetted surfaces are handled. Avoiding the use of plastics in the laboratory can best minimize interferences from phthalates. Matrix interferences may be caused by contaminants that are co-extracted from the sample. The cleanup procedures described in the Preparation SOP can be used to overcome many of these interferences. For more details see Glassware Cleaning Protocol in the Organic Preparations Laboratory DEP SOP GC-003-2 Ref . 15.5.

4. DEFINITIONS

4.1. ACCURACY: The closeness of agreement between an observed value and an accepted reference value. When applied to a set of observed values, accuracy will be a combination of a random component and of a common systematic error (or bias) component.

4.2. BATCH: A group of samples, which behave similarly with respect to the sampling or the testing procedures being employed, and which are processed as a unit. For QC purposes, if the number of samples in a group is greater than 20, then each group of 20 samples or less will all be handled as a separate batch.

4.3. BIAS: The deviation due to matrix effects of the measured value (Xs - Xu) from a known spiked amount. Bias can be assessed by comparing a measured value to an accepted reference value in a sample of known concentration or by determining the recovery of a known amount of contaminant spiked into a sample (matrix spike). Thus, the bias (B) due to matrix effects based on a matrix spike is calculated as:

$B = (Xs - Xu) - K$
Where:
Xs = measured value for spiked sample,
Xu = measured value for unspiked sample, and
K = known value of the spike in the sample.

Using the following equation yields the percent recovery (%R).

$\%R = 100 (Xs- Xu)/ K$

4.4. PRACTICAL QUANTITATION LIMIT (PQL): The lowest concentration that can be reliably achieved within specified limits of precision and accuracy during routine laboratory operating conditions. The PQL is generally 5 to 10 times the MDL. However, it may be nominally chosen within these guidelines to simplify data reporting. For many analytes the PQL analyte concentration is selected as the lowest non-zero standard in the calibration curve. Sample PQLs are highly matrix-dependent.

4.5. FIELD DUPLICATES: Independent samples which are collected as close as possible to the same point in space and time. They are two separate samples taken from the same source, stored in separate containers, and analyzed independently. These duplicates are useful in documenting the precision of the sampling process.

4.6. LABORATORY FORTIFIED BLANK (LFB): A known matrix spiked with compound(s) representative of the target analytes. The LFB is analyzed exactly as a sample. This is used

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to determine whether the methodology is in control, and whether the laboratory is capable of making accurate and precise measurements.

4.7. MATRIX: The component or substrate (e.g., surface water, drinking water), which may contain the analyte of interest.

4.8. MATRIX SPIKE: An aliquot of sample spiked with a known concentration of target analyte(s). The spiking occurs prior to sample preparation and analysis. A matrix spike is used to document the bias of a method in a given sample matrix.

4.9. MATRIX SPIKE DUPLICATES: A second aliquot of the same sample used for matrix spike spiked with identical concentrations of target analyte(s). The spiking occurs prior to sample preparation and analysis. They are used to document the precision and bias of a method in a given sample matrix.

4.10. METHOD BLANK: An analyte-free matrix to which all reagents are added in the same volumes or proportions as used in sample processing. The method blank must be carried through the complete sample preparation and analytical procedure. The method blank is used to document contamination resulting from the analytical process. For a method blank to be acceptable for use with the accompanying samples, the concentration in the blank of any analyte of concern must not be higher than the method detection limit.

4.11. METHOD DETECTION LIMIT (MDL): The minimum concentration of a substance that can be measured and reported with 99% confidence that the analyte concentration is greater than zero and is determined from analysis of a sample in a given matrix type containing the analyte. For operational purposes, when it is necessary to determine the MDL in the matrix, the MDL should be determined by multiplying the appropriate one-sided 99% t-statistic by the standard deviation obtained from a minimum of seven analyses of a matrix spike containing the analyte of interest at a concentration one to five times the estimated MDL.

4.12. ORGANIC-FREE REAGENT WATER: All references to water in the method refer to water in which interference is not observed at the method detection limit of the compounds of interest. Passing tap water through a carbon filter bed containing about 1 pound of activated carbon can generate organic-free reagent water. A water purification system may be used to generate organic-free deionized water.

4.13. PRECISION: The agreement among a set of replicate measurements analyses. These samples should contain concentrations of analyte above the MDL, and involve the use of matrix spikes or laboratory fortified blanks. The estimates of precision are based on the relative without assumption of knowledge of the true value. Precision is estimated by means of duplicate percent difference (RPD).

$$RPD\ (\%) = 100 \times 2\ (X1 - X2) / (X1 + X2)$$

where, X1 and X2 are the two measurements

4.14. QUALITY CONTROL CHECK STANDARD (QCS): Standard solution from a source, other than normal calibration standards, that is certified and traceable. These standards are used to check the accuracy of a calibration curve.

4.15. SPIKE SOLUTION (SS): A solution of method analytes of known concentrations that is used to fortify an aliquot of laboratory reagent water or sample matrix.

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4.16. STANDARD CURVE: A plot of concentrations of known analyte standards versus the instrument response to the analyte. Calibration standards are prepared by successively diluting a standard solution to produce working standards that cover the working range of the instrument. Standards should be prepared at the frequency specified in section 6. The calibration standards should be prepared using the same type of solvent as will result in the samples following sample preparation.

4.17. SURROGATE: An organic compound which is similar to the target analyte(s) in chemical composition and behavior in the analytical process, but which is not normally found in environmental samples used to monitor the preparation. Surrogates are added to each field and QC sample prior to extraction.

5. APPARATUS AND EQUIPMENT

5.1. Gas Chromatograph: HP 6890 or Agilent 7890A capable of four steps temperature programming, with dual electron capture detectors, dual capillary split/splitless injectors, and a dual HP 7683A and 7683B Automatic Samplers. The GCs and GC Automatic Samplers are set at the operating parameters shown in Table 1 and Table 2.

5.1.1. First analytical column is a DB-1701, J & W fused silica capillary, 60 m x 0.25 mm x 0.25 um or similar column.

5.1.2. The second column is a DB-5, J & W fused silica capillary, 60 m x 0.25 mm x 0.25 um or similar column.

5.2. Data system

5.2.1. The EZChrom Elite Client/Server software provided by Agilent Technologies, Inc. is interfaced to the Chromatographs, which allows the continuous acquisition and storage on machine-readable media of all chromatographic measurements obtained throughout the duration of the chromatographic run sequence program.

5.2.2. The processing and uploading of the chromatographic data to LIMS is performed using the Target software provided by Thermo Fisher Scientific, Inc.

5.2.3. For reporting data, the system currently uses the FDEP LIMS software.

5.3. Gases

5.3.1. Helium, UPC Grade, is used as the carrier gas. The helium is passed through oxygen and hydrocarbon filters before it enters the GC.

5.3.2. Nitrogen, UPC Grade, is used as the make up gas for the ECD detectors. The N_2 is passed through oxygen and hydrocarbon filters before entering the GC.

5.3.3. Gases are delivered to the instruments from a Laboratory Gas Manifold System.

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6. REAGENTS AND CHEMICALS

6.1. Solvents

6.1.1. ISO-Octane, Fisher Scientific, Pesticide, HPLC or OPTIMA grade.

6.1.2. Acetone, Fisher Scientific, Pesticide, HPLC or OPTIMA grade.

6.2. Stock and Intermediate Standards

6.2.1. Intermediate Standards mixtures are also prepared from stock standards once per year by diluting with iso-octane or other appropriate solvents. Stock standards may be prepared in house or purchased from a certified manufacturer (ISO 9000 or EPA certified).

6.2.2. Intermediate standard solutions are mixtures or individual standards used to prepare working standards. There are five standard mixtures used in this procedure: ECD A, ECD F, ECD C, CONGENERS MIXTURE, and TOXAPHENE. EDC C is used to monitor the breakdown of DDT and Endrin. New standards are checked vs. a second source (QC Standards), after diluting them to one of the working standard levels. The acceptance criterion is +/- 15 %. All standard preparations and verifications have to be recorded in the Standard Preparation Tracker located in FL DEP LIMS. A list containing all standards with their serial numbers used in the analysis has to be part of analysis documentation folder.

6.3. Working Standard Solutions (Calibration standards)

6.3.1. Working standard solutions are a series of diluted intermediate standards in iso-octane. They are used for calibration.

6.3.2. The concentrations for each analyte in the intermediate and diluted standards are listed in Table 4.

6.4. Spiking Solution

6.4.1. These solutions are used when fortifying sample matrix spikes or lab fortified blanks at extraction. These fortified samples are used to verify the accuracy and precision of the sample preparation and analysis.

6.4.2. Spike solutions are prepared by diluting stock or intermediate standards with acetone. Concentrations of the parameters in these solutions are listed in Table 4.

6.4.3. Freshly prepared spike solutions are checked vs. valid working standards. The acceptance criterion is +/- 10 %.

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6.5. Quality Control Check Standards (QCS)

6.5.1. Quality Control Check Standards are prepared from a source, other than calibration standards, that is certified and traceable. They are usually prepared at PQL or middle calibration level. These standards are used to check the integrity of the calibration standards and are run and analyzed whenever full range initial calibration curve is built. The acceptance criterion is +/- 15% difference.

6.6. Look into Appendix 1 for detailed standard preparation.

7. SAMPLE COLLECTION, PRESERVATION, AND HANDLING

7.1. Samples must be collected in glass jars with Teflon-lined screw caps.

7.2 Please refer to DEP SOP GC-00401 for sample preservation.(Ref 15.1) 7.3 Samples must be extracted within 365 days of collection and completely analyzed within 40 days of extraction.

8. SAMPLE PREPARATION PROCEDURE

8.1. For preparation procedure see DEP SOP, GC-004-1(Ref 15.1,): "EXTRACTION OF ORGANOPHOSPHORUS, ORGANONITROGEN AND ORGANOHALIDE PESTICIDES and PCBs, FROM FISH TISSUE FOR SUBSEQUENT ANALYSIS BY GC-NPD/ECD".

9. GENERAL DESCRIPTION OF GC/ECD ANALYSIS

9.1. Pre-Run Equipment Maintenance

9.1.1. Before starting an analysis run sequence, an initial checking and maintenance of the instrument has to be performed.

9.1.2. Initial checking and maintenance of the instrument consists mainly in the following steps:

9.1.2.1. Check the detector signals to be in the accepted range

9.1.2.2. Run and analyze an Endrin/DDT mixture standard and determine the % decomposition

9.1.2.3. If %decompositions exceed the accepted limit of 15% for any of these components then:

9.1.2.4. Replace the injector liner, cut 3-5 inches of the column, and replace the injector gold seal.

9.1.2.5. Re-determine the % decomposition and make sure that is below the accepted limit

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9.1.2.6. Run and Instrument Blank and look for Instrument Contamination.

9.1.2.7. In case the instrument is contaminated raise the furnace temperature to 260 ^{o}C and leave it overnight.

9.1.2.8. Re-check for Instrument contamination and if needed repeat the procedure until the instrument is contamination free.

9.2. Summary of GC/ECD Analysis

9.2.1. The analysis of samples is accomplished by using two dissimilar fused silica capillary columns, usually a DB-5 and a DB-1701 or similar column (usually 0.25mm ID/60m/0.25um

9.2.2. One of the two columns, based on the best performance, is chosen by the analyst to be “primary”. The “primary column” is used to report the results. Important criteria for choosing the “primary column”:

9.2.2.1. Stable retention time windows during the entire run sequence

9.2.2.2. The best chromatography (no tailing or fronting, no impurity interference with surrogates of target components)

9.2.2.3. The best calibration curves especially for target positives

9.2.2.4. The best LCS/Matrix Spikes results

9.2.2.5. The smallest number of CCV failures

9.2.2.6. The best surrogates results

9.2.2.7. The lowest %decomposition for Endrin/DDT

9.2.3. Analysis is accomplished in two steps: identification of positives and quantification of the positives.

9.2.4. Identification of positives requires all of the following quality criteria to be met:

9.2.4.1. Use of an updated single through origin (for Toxaphene), or multi-point calibration (updated Retention Times and Response Factors) for all single peak components.

9.2.4.2. Stable retention time windows (all calibration check standards are within the calibration windows).

9.2.4.3. Recoveries for surrogates and spikes are within the control limits.

9.2.5. All samples are analyzed in two columns for confirmation of the positives. The LFBs, Matrix Spikes, and positive reported results, are based on the “primary column” (chosen by the analyst). If for some analytes QC criteria in the primary column are out of acceptable range, due to matrix interference or other

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problems, the results will be reported using the values found in the confirmatory column if the QC criteria are within limits.

9.2.6. A single peak target component is identified as positive if its peak is detected within its retention time window in both columns and the response exceeds the MDL. A multiple peak target component like Toxaphene, is identified primarily by pattern recognition although significant modifications are expected for tissue matrix. The amount of the prospective positive found in one column has to be confirmed by the amount found in the other column. Criterion for the amount confirmation is equal to or less than +/- 30% difference between the two columns values. For Toxaphene this criterion is equal to or less than +/- 50%. If besides retention time, there is other evidence that the target analyte is a positive, and the amount in two columns does not match, the result will be reported based on the smallest value found in the two columns, and will be qualified as estimated (J).

9.2.7. Quantitative analysis is accomplished by the external calibration mode. The calibration fits used are Average Response Factor, linear or quadratic. For building an initial calibration curve a minimum of 5 calibration levels must be used. Six levels are required for quadratic fits. The linearity of the calibration curve is defined by an RSD of 20 % or less of the Average Response Factors, or by a Correlation Coefficient of 0.995 or more. After identification, all positives are quantified. If the quantity found is not within the calibration range the samples are diluted or re-run with a multilevel calibration curve.

9.2.8. The analyst should monitor the performance of the extraction and cleanup procedures by spiking each sample, QC sample and Method Blank with pesticide surrogates. Because GC/ECD analysis is subject to interferences, two surrogates are to be used. Decachlorobiphenyl (DCB) is the primary surrogate, and should be used whenever possible. However, if recovery for DCB fails, then TMX (2,4,5,6-tetrachloro-m-xylene) should be evaluated for acceptance (See Ref 15.3). To determine acceptance accuracy limits for surrogates the following procedure should be used (Ref 15.6): (1) for each sample analyzed, calculate the percent recovery of each surrogate in the sample, (2) once a minimum of thirty samples of the same matrix has been analyzed, calculate the average percent recovery (p)and standard deviation of the percent recoveries (s) for each of the surrogates, (3) for a given matrix, calculate upper and lower control limit for method performance for each surrogate standard as follows:

Upper Control Limit (UCL) = p + ts
Lower Control Limit (LCL) = p - ts

Where “t” stands for Student’s coefficient for n-1 degrees of freedom (n=number of measurements).

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Control Limits are checked once per year. The statistical control limits can be replaced by statutory control limits if they are practically comparable.

9.2.9. If surrogate recoveries are outside control limits, the sample has to be re-run, and reanalyzed. If surrogates fail again sample must be re-extracted and re-analyzed. If the sample exceeded the holding time for extraction, and the recovery is biased high, the re-extraction is not required and the reported positive hits have to be qualified as estimated with the following comment entered in the Sample Report: “Estimated value for 'parameter' is due to surrogate recovery for TMX/DCB out of control limits”. If surrogates are missing or the recovery is biased low, samples should be re-extracted and reanalyzed even if they are beyond holding time.

9.2.10. Absolute Retention time windows are used for the identification of target analytes. The absolute retention time window is assessed during initial calibration as +/- 0.05 minute of the mean RT of the analyte.

9.2.11. The decomposition of DDT and Endrin is evaluated by injecting an ECD C (Endrin/DDT decomposition) standard (containing Endrin at 160 ug/L and DDT-p,p’ at 100 ug/L) at the beginning and at least every 12 hours afterwards. Presence of DDE, DDD, Endrin Aldehyde and Endrin Ketone indicates breakdown. Before starting a run sequence maintain the instrument until the breakdown is less than 15 % in both columns for both DDT and Endrin. The ECD C chromatograms are analyzed using “% Dec. Target sublist”. Calculate the breakdown as follow:

%Dec. DDT= Sum Peak Area (DDE+DDD)/Sum Peak Area (DDE+DDD+DDT) x 100
%Dec. Endrin = Sum Peak Area (En.Ald. + En.Ket.)/Sum Peak Area (En.Ald. + En.Ket + Endrin) x 100

“If the 12 hours interval (beyond the initial) Endrin/DDT decomposition check fails (exceeds 15%), rerun and reanalyze those samples in which DDT and /or Endrin and their decomposition products were detected positive. If the samples are beyond holding time, flag the results as estimated and include appropriate comments.”

9.2.12. To demonstrate that interferences from the analytical system , reagents and glassware are under control, the analyst must analyze one Instrument Blank (clean Iso-octane) in the beginning of each run sequence and one Method Blank (extraction blank) for each extraction batch. A blank will be within acceptable range, if the concentration in that blank of any analyte of interest is below the analyte MDL.

9.2.13 In case a positive identification is found in both the sample(s) and the extraction blank then the sample batch has to be re-extracted and reanalyzed. If samples passed the holding time the affected samples’ results will be reported with a “V” qualifier up to ten times the amount found in the blank.

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9.3. Instrument Initial Calibration

9.3.1. Quantitation of a positive hit at PQL or higher level for all target components is accomplished by using most commonly linear or quadratic fit of the calibration curve. To create an initial calibration curve using external standard procedure at least five (six for quadratic fits) standard calibration levels should be used. One of the calibration levels must be at a concentration lower than or equal to the PQL, but higher than MDL. The other concentrations should correspond to a range of concentrations for which an acceptable linear/quadratic calibration curve can be drawn. The Chromatography Software will calculate the ratio of response to the amount injected (calibration factor).

9.3.2 The system is considered to be in initial linear calibration if the Relative Standard Deviation (%RSD) of the analyte calibration factors (using peak area calculation) is less or equal to 20% or the Correlation Coefficient is not smaller than 0.995. Linear fit is chosen as a first choice for the calibration curve fit in Target software, followed by average response factor (mathematically equivalent to weighted linear) and then followed by quadratic fit. Whenever quadratic fit is applied at least six calibration standards are required. The calibration curve is additionally checked by re-fitting the lowest calibration point back to the model. The calculated amounts for each of the standard components should be within 20% of the expected values.

9.3.3 All initial calibration curves shall be checked vs. middle range QCS standard and %Recovery must be within +/- 15%. If this Quality Criterion fails for parameters reported as positives, a new calibration curve must be built, samples re-run and reanalyzed. If the samples are expired, affected results must be reported as estimated (J).

9.4. Calibration Verification

9.4.1. After initial calibration, the detector responses for all parameters are checked on a continuing basis, at least once per run sequence (routinely every 7-10 samples) by using a continuing calibration verification standard (CCV). Criterion of acceptance for continuing calibration verification is +/- 15 % of initial calibration response. The channel with acceptable CCV checks will be used to quantify and report the positives if all other quality criteria are met. If CCV fails in both channels, all samples bracketed by this standard have to be re-run for all identified positive hits exceeding PQL level. If the samples are beyond analysis holding time, all reported positive hits with failing CCV must be flagged as estimated (J).

9.4.2. If CCVs do not meet method requirements the associated analytical results must be qualified in accordance with Lab SOP CM-008-1 (Ref 15.14).

9.5. Analysis Run Sequence.

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9.5.1. The analyst will set up an analysis run sequence by placing the GC vials containing the samples extracts and standards on the GC auto sampler tray in the following order or similar:

- Iso-octane blank (equipment blank)
- Endrin/DDT decomposition Check.
- ECDA L1
- ECD A L2
- ECD A L3
- ECD A L4
- ECD A L5
- ECD A L6
- ECD A QCS
- CNGPCB-L1
- CNGPCB-L2
- CNGPCB-L3
- CNGPCB-L4
- CNGPCB-L5
- CNGPCB-L6
- CNGPCB QCS
- ECDC+F-L1
- ECDC+F-L2
- ECDC+F-L3
- ECDC+F-L4
- ECDC+F-L5
- ECDC+F-L6
- ECDC+F-QCS/
- TOXA-L3
- ECDA+C+F-L3 CCV
- CONGPCB-L3 CCV
- Endrin/DDT decomposition Check.
- DE BLK (Diatomeceaous Earth blank)
- MBLK
- LCS
- MS
- MSD
- GPCBLK1
- GPCBLK2
- SAMPLE #1 to #7
- EDC A-L3 CCV
- ECD C+F-L3 CCV
- CNGPCB-L3 CCV
- TOXA-L3 QCS/CCV
- Endrin/DDT decompositiom Check.
- SAMPLE #7 to 17
- ALL CCVs
- Etc.

9.6. Data Processing/Reporting

9.6.1. Identification

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9.6.1.1. Analytes are identified by matching retention times with those that are in the standard chromatographs. For Toxaphene the identification is primarily based on pattern recognition.

9.6.2. Quantification

9.6.2.1. Positives are quantified based on their peak area. The concentration reported for an identified hit in the sample extract, expressed in ug/L, is based on calibration curve of the initial calibration. The Target software, used to process raw data, will do the interpolation for the sample extract. However, the analyst must ensure that all identification criteria are fulfilled and the peak area integration was correctly done. If not, the integration has to be corrected by choosing a new set of integration parameters or by manual integration (for manual integration procedure see Ref 15.8). Additionally, Target will complete the calculation to determine the concentration of the analyte in the sample. This is done as follows:

C_{sample} (ug/kg) = $(C_x) \times (V_{extract}) \times (DF)/(W_{sample})$

Where:

C_x – the concentration, in ug/L, of the analyte in the sample extract.

$V_{extract}$ – the volume of the extract, in mL (usually 4 mL) recorded in the preparation report

DF – the appropriate dilution recorded in the dilution report

W_{sample} – wet sample weight, expressed in grams recorded in the preparation report

9.6.2.2. For Toxaphene, which is a multiple peak target component, the quantification is significantly more complicated than that of single component parameters. The GC pattern of a Toxaphene residue may differ considerably from that of the calibration standard. Depending on the sample history, residue of Toxaphene can consist of any combination of constituents from Technical Toxaphene standard and fish digestion metabolites, and products of degradation caused by exposure to environmental factors such as water or sunlight. Fish exposed to a recent spill of Technical Toxaphene will contain a residue drastically different from a fish whose Toxaphene residue was accumulated by ingestion of smaller fish or vegetation. Due to this inability to predict a Toxaphene GC pattern, it is not possible to prescribe a single method for quantification. The analyst can chose between “Peak method” and “Total area method” depending on which is more appropriate for the sample.

9.6.2.3. In the “Peak method” approach, the analyst chooses 5-7 predominate peaks covering the whole elution range of the Toxaphene standard equally. These peaks are not supposed to co-elute with other targeted

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chlorinated parameters or other unidentifiable matrix peaks. This approach is to be considered whenever the GC pattern in the sample and standard are similar, or when the sample matrix or other contaminants will significantly interfere with the Toxaphene GC pattern.

9.6.2.4. In the "Total area method" approach, the analyst chooses the total area of the GC Toxaphene standard chromatogram for calibration and quantification. This method is the best choice when Toxaphene is the major contaminant, and in the case of no significant matrix interferences. This method may require subtraction of major interfering peaks.

9.7. Reporting results in FL DEP LIMS.

9.7.1. QA/QC results, MDLs, and positives are reported in the DEP LIMS using the Target command: "Custom Functions / Upload to DEP QCM". QC Manager Instrument: GC_ECD_5.(or other GC_ECD_#).

10. QUALITY CONTROL

10.1. For QA/QC see the Lab SOP GC-001-2 (Ref 15-9).

10.2. QC criteria pertaining to this SOP are summarized in Table 7

10.3. All data generated by a chemistry analyst are subjected to supervisory review and authorization. . The supervisor is assessing the validity of data generated by assuring that all QA/QC criteria are met, making use of the following review/authorization checklist: (See also Lab SOP OG-003-2.)

- Check Sample preparation report
  - Procedure
  - Surrogate and spiking solutions
  - Preparation holding time
- Check customer special request.
- Check Initial calibration for quality criteria
- Initial calibration for second source QCS standard
- Check continuing calibration verification (CCV) standards
- Check surrogate recoveries
- Check Accuracy and precision for LFB and Matrix Spikes
- Check QCS recoveries
- Check Retention time drift
- Check Decomposition for Endrin / DDT
- Check Blanks contamination
- Check for positives:
  - Integration of the peaks (baseline)
  - Retention time confirmation
  - Amount confirmation
  - If response is in the calibration range
  - Calculations
  - Qualifiers

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- Check information in the Target report header:
  - Instrument ID
  - Column ID
  - SOP # and Instrument Serial #
- Evaluate the data against any field information
- Check LIMS report:
  - Preparation and analysis dates
  - MDLs
  - Amounts reported for positives
  - Qualifiers
  - Comments
  - CCV reports
  - Method BLK report
  - Surrogate report
  - For more detail see SOP OG-003-2, Reference 15.16

11. METHOD PERFORMANCE

11.1. The MDLs are determined by using EPA recommended procedure (40 CFR Pt 136 Appx. B) (Ref15.10), by analyzing at least 7 LCS spikes in reagent tissue matrix or if not available, sodium sulfate at 1-5 times the estimated MDL level. The spikes are run and analyzed in DB-5 / DB- 1701. The Reported MDLs are chosen based on the highest value measured in the two columns. The equation used to calculate the MDL is as follows:

$$MDL = S \times t_{(n-1,\ 1-a=0,99)}$$

where: $t_{(n-1,\ 1-a=0.99)}$ is Student Coefficient for the 99% confidence level for
n= number of replicates
S= standard deviation

MDLs must be verified annually, for each analyte.

12. DATA ARCHIVAL

12.1. All raw data are retained in electronic and hard copy format. All paper forms are maintained either in the laboratory office or in a department of State warehouse for ten years. Electronic chromatographic raw data are archived by analysis file ID on duplicate CDs by the Laboratory Support Section.

13. SAFETY

13.1. The toxicity or carcinogenic nature of each reagent used in this method has not been precisely defined; however, each chemical compound should be treated as a potential health hazard. From this viewpoint, exposure to these chemicals must be reduced to the lowest possible level by whatever means available. The laboratory is responsible for maintaining a current awareness file of OSHA regulations regarding the safe handling of the chemicals specified in this method. A reference file of material data handling sheets should also be made available to all personnel involved in the chemical analysis. The following parameters covered by this method have been tentatively classified as known or suspected, human or mammalian carcinogens: 4,4-DDT, 4,4-DDD, the BHCs, and the PCBs. 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.

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13.2. Lab clothing

13.2.1. Lab coats and safety eyeglasses must be worn at all times when in any laboratory. Use gloves when handling samples, standards, or solvents, or when washing glassware.

13.3. Chemicals involved

13.3.1. Even though the toxicity or carcinogenic nature of some of the reagents used in this method has not been precisely defined, each of them should be treated as a potential health hazard. From this point of view exposure to these chemicals must be reduced to the lowest possible level by whatever means available.

13.3.2. A reference file of Material Safety Data Sheets for each reagents used, is kept in front of the tech room (room A316J) for documentation. Refer to this folder for any materials you are handling. Update this folder upon receiving new chemicals or reagents.

13.3.3. Primary standards of these toxic compounds should be prepared in a hood (See "Hood Usage"). A NIOSH/MESA approved toxic gas respirator should be worn when handling highly concentrated solutions of these compounds.

13.3.4. Flammable solvents must be stored in a flammables cabinet.

13.4. Waste disposal (see Lab Manual "Laboratory Waste Management" January 2007)

13.4.1. Neat standards should be sealed and labeled. All stock standards, working standards, and unused sample extracts must be emptied into two waste containers separately identified for Pesticides and PCBs and labeled properly: Pesticides waste and PCBs waste.

13.4.2. Chlorinated and non-chlorinated solvents must be stored separately and labeled properly.

13.4.3. GC vials containing, standards, and sample extracts, must be stored in two separate containers for Pesticides and PCBs. They should be stored in the hood and labeled properly: Pesticides waste and PCBs waste.

13.5. Hood Usage

13.5.1. Handle all sample extractions, solvent transfers, concentrations, sample preps, and clean-ups in the hood at all times.

13.5.2. At any time, no more than two people can work at each hood.

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13.5.3. Turn the hood on and wait for adequate airflow (If no airflow, DON'T USE!).

13.6. Glassware

13.6.1. Inspect every piece of glassware to be used. Do not use any that are chipped, cracked, etched, or scratched.

13.6.2. Dispose of all unwanted, broken glassware in the blue "GLASS" boxes provided. One should discriminate between "unwanted broken" glassware and that which is only slightly damaged and reasonably easily repairable.

13.7. For more details see DEP SOP OG-001-3 (Ref 15.11)

14. POLLUTION PREVENTION

14.1. Standards, QC Samples and Extracts of contaminated samples are disposed following laboratory protocol for hazardous waste disposal SOP OG-001-3 (Ref.15-9).

14.2. Contaminated samples and all other disposed chemical wastes are sent to specialized company for incineration.

15. REFERENCES

15.1. DEP SOP GC-004-1 Extraction of Organophosphorus, Organonitrogen, Organohalide Pesticides and PCBs from Fish Tissue for Subsequent Analysis by GC-NPD/ ECD.

15.2. EPA 8081B Organochlorine Pesticides by Gas Chromatography; Test Methods for Evaluating Solid Wastes, Physical/Chemical Methods, SW 846; 3rd Edition, Update 4, January 3, 2008.

15.3. EPA 8082A Polychorinated Biphenys (PCBs) by Gas Chromatography; Test Methods for Evaluating Solid Wastes, Physical/Chemical Methods, SW 846; 3rd Edition, Update 4, January 3, 2008.

15.4. M. Schantz, R. Parris, J. Kurtz, K. Ballschmiter and S.Wise Fresenius J. Chem. 345,766-778 (1993).

15.5. DEP SOP CM-0031-1 Glassware Cleaning Protocol in the Organic Preparations Laboratory

15.6. EPA 8000C Determinative Chromatographic Separation; Test Methods for Evaluating Solid Wastes, Physical/Chemical Methods, SW 846; 3rd Edition, Update 4, January 3, 2008.

15.7. DEP SOP CM-008-1 Standard Operating Procedure for Reporting Qualified Data.

15.8. DEP SOP CM-018-1 Laboratory Policy Regarding Manual Chromatographic Peak Integration

15.9. DEP SOP GC-001-2 Quality Assurance/Quality Control in the GC Pesticides Laboratory.

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15.10. 40 C.F.R. Appendix B to Part 136; Definition and Procedure for the Determination of the Method Detection Limit-Revision 1.11.

15.11. DEP SOP OG-001-3; Safety and Waste Management Practices in the Organic Analysis Groups (Volatile, Semi-Volatile and Pesticide Laboratories)

16. APPENDIX 1

Standard Preparation Procedure

Stock solutions usually come in 1mL glass vials. Take the vials out of the freezer at least 30min before preparation. Open vials and transfer the entire solution into 10mL centrifuge vials (using Pasteur pipettes). Record the transfer date and keep the vial label.

The Intermediate Standard is prepared by diluting the Stock solution. The Working Level Standards are prepared by diluting the Intermediate Standard.

Surrogates:
The individual surrogate stock solutions are prepared from purchased neat material.

Neat	Components	Source
100mg	TMX	Chem Service
100mg	DCB	Chem Service

Individual Stock solutions:
Solvent: Acetone
Note: DCB does not dissolve easily. Use the heated ultrasonic bath for 1-2 h.

Component	Neat added mg	Stock V_f mL	Conc. Stock ug/mL
TMX	10	50	200
DCB	10	50	200

Surrogate spike solution (Mix):
Solvent: Acetone

Component	Amount Stock added mL	Dilution Factor	Surr. spike V_f mL	conc.ug/mL
TMX*	3.125	80	250 ml	2.5
DCB**	6.250	40		5
NMX***	12.5	20		50

*Tetrachloro-M-Xylene.
**Decachlorobiphenyl.
***Nitro-M-Xylene.

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ECDA Custom Mix:

Solvent: Acetone

Stock conc. ug/mL	Components	Source
100	Aldrin, Alpha-BHC, Alpha-Chlordane, Beta-BHC, Delta-BHC, Dieldrin, Endosulfan I, Endosulfan II, Gamma-BHC, Gamma-Chlordane, Heptachlor, Heptachlor Epoxide,	1.Ultra Scientific 2.Accu Standard
200	DDD-p,p', DDE-p,p', Endosulfan Sulfate, Endrin Aldehyde, Endrin Ketone,	
500	Methoxychlor	

Intermediate ECDA Standard Solution:

Amount **Ultra Scientific ECDA** Stock added **mL**	Dilution Factor	**INT** in acetone **V_f mL**	conc. ug/mL
1	100	**100**	1
			2
			5
Amount **TMX** Stock added (mL)			
0.5	200		1
Amount **DCB** Stock added (mL)			
1	100		2

Amount **Accu Standard ECDA** Stock added mL	Dilution Factor	**INT QCS** in ISO-8 **V_f mL**	conc. ug/mL
1	100	**100**	1
			2
			5

ECDA Working Level Standards:

Solvent: ISO-8

	V_f mL	**Amount ECDA INT added mL**	**Amount ECDA QCS INT added mL**	**Dilution Factor**	**Conc. ug/L**
L1	**100**	**5**	**--**	**20**	**50**
					100
					250

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L2	**200**	**7.5**	**--**	**26.666**	**37.5**
					75
					187.5
L3	**200**	**5**	**--**	**40**	**25**
					50
					125
L4	**200**	**2**	**--**	**100**	**10**
					20
					50
L5	**100**	**0.4**	**--**	**250**	**4**
					8
					20
L6	**100**	**0.2**		**500**	**2**
					2
					4
					10
L3 QCS	**200**		**5**	**40**	**25**
					50
					125

ECDC Custom Mix:

Solvent: Acetone

Stock conc. ug/mL	Components	Source
250	DDT-p,p'	1.Crescent Chem. 2.Accu Standard
250	Endrin	

Intermediate ECDC Standard Solution:

Amount **Ultra Scientific ECDC** Stock added **mL**	Dilution Factor	**INT** in acetone	
		V_f mL	conc. ug/mL
1	100		2.5
		100	2.5
Amount **Accu Standard ECDC** Stock added (mL)	Dilution Factor	**INT QCS** in acetone	
		V_f mL	conc. ug/mL
1	100	**100**	2.5
			2.5

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ECDC Working Level Standards:
Solvent Working Level Standards: ISO-8 Spike: 50:50 Acetone: ISO-8

	V_f mL	Amount ECDC INT added mL	Amount ECDC QCS INT added mL	Amount ECDA QCS INT added mL	Dilution Factor	Conc. ECDC ug/L	Conc. ECDC ug/L
L1	**100**	**4**	**--**	**--**	**25**	**100** **100**	**--**
L2	**100**	**3**	**--**	**--**	**33.333**	**75** **75**	**--**
L3	**200**	**4**	**--**	**--**	**50**	**50** **50**	**--**
L4	**250**	**2**	--	**--**	**125**	**20** **20**	**--**
L5	**250**	**1**	--	**--**	**250**	**10** **10**	**--**
L6	**100**	**0.32**			**312.5**	**8** **8**	
L3 QCS	**200**	**--**	**4**	**--**	**50**	**50** **50**	**--**
ECDA/C spk	**250**	**--**	**20**	**25**	**12.5** **10**	**200** **200**	**100** **200** **500**
ECDC MDL	**250**	**0.25**	**--**	**--**	**1000**	**2.5** **5**	**--**

ECDF Custom Mix:
Solvent: Acetone

Stock conc. ug/mL	Components	Source
100	Hexachlorobenzene, Trans-Nonachlor, Cis-Nonachlor	1.Crescent Chem. 2.Accu Standard
200	DDT-o, p', DDD-o,p', DDE-o,p', Mirex	

Intermediate ECDF Standard Solution:

Amount **Ultra Scientific ECDF** Stock added (**mL)**	Dilution Factor	**INT** in acetone **V_f mL**	conc. ug/mL
1	100	**100**	1 2
Amount **Accu Standard ECDF** Stock added mL	Dilution Factor	**INT QCS** in aceton **V_f mL**	conc. ug/mL
1	100	**100**	1 2

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ECDF Working Level Standards:

Solvent Working Level Standards: ISO-8 Spike: Acetone

	V_f (mL)	Amount ECDF INT added (mL)	Amount ECDF QCS INT added (mL)	Dilution Factor	ECDF Conc. (ug/L)
L1	**100**	**5**	**--**	**20**	**50** **100**
L2	**200**	**7.5**	**--**	**26.666**	**37.5** **75**
L3	**100**	**2.5**	**--**	**40**	**25** **50**
L4	**100**	**1**	**--**	**100**	**10** **20**
L5	**100**	**0.4**	**--**	**250**	**4** **8**
L6	**100**	**0.2**		**500**	**2** **4**
L3 QCS	**100**	**--**	**2.5**	**40**	**25** **50**
ECDF spk	**100**	**--**	**10**	**10**	**100** **200**

Toxaphene Stock Standard Solution:

Solvent: Acetone

Stock conc. ug/mL	Components	Source
1000	Toxaphene	1. Accu Standard 2.Accu Standard

Toxaphene Intermediate Standard Solution:

Amount **Ultra Scientific TOXA** Stock added **mL**	Dilution Factor	**INT** in acetone	
		V_f mL	conc. ug/mL
1	100	**100**	10
Amount **Accu Standard TOXA** Stock added mL	Dilution Factor	**INT QCS** in acetone	
		V_f mL	conc. ug/mL
1	100	**100**	10

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Toxaphene Working Level Standards:

Solvent Working Level Standards: ISO-8 Spike: Acetone

	V_f mL	Amount TOXA INT added mL	Amount TOXA QCS INT added mL	Dilution Factor	Conc. TOXA ug/L
L1	**50**	**25**	**--**	**2**	**5000**
L2	**50**	**10**	**--**	**5**	**2000**
L3	**100**	**10**	**--**	**10**	**1000**
L4	**50**	**3**	**--**	**16.666**	**600**
L5	**50**	**1.5**	**--**	**33.333**	**300**
L6	**50**	**1**	**--**	**50**	**200**
L3 QCS	**100**	**--**	**10**	**10**	**1000**
TOXA spk	**INT QCS used as spike**	**--**			**10000**

PCB Congener Mix :
Solvent: Acetone

Stock conc. ug/mL	Components	Source
100	PCB 8, PCB 18, PCB 28, PCB 44, PCB 52, PCB 66, PCB 77, PCB 101, PCB 105, PCB 118, PCB 126, PCB 128, PCB 138, PCB 153, PCB 170, PCB 180, PCB 187, PCB 195, PCB 206,	1.Crescent 2.Accu Standard

PCB Congener Intermediate Standard Solution: Prepared by mixing PCB congener stock solution and DDT-o,p' stock solution.

Amount **Crescent Congener Mix** Stock added **mL**	Dilution Factor	**INT PCB congener** in acetone	
		V_f **mL**	conc. ug/mL
1	100	**100**	1

Amount **Accu Standard Congener Mix** Stock added **mL**	Dilution Factor	**INT QCS PCB congener** in acetone	
		V_f **mL**	conc. ug/mL
1	100	**100**	1

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PCB Congener Working Level Standards:

Solvent: ISO-8 Spike: Acetone

	V_f mL	Amount PCB congener INT added mL	Amount PCB congener QCS INT added mL	Dilution Factor	Conc. ug/L
L1	100	10	--	10	100
					200
L2	100	7.5	--	13.33	75
					150
L3	100	5	--	20	50
					100
L4	100	2.5	--	40	25
					50
L5	100	1	--	100	10
					20
L6	100	0.5	--	200	5
					10
L3QCS	100	--	5	20	50
					100
PCB congener spk	100	8 --	10	10	100
					200

17. TABLES

TABLE 1: GC OPERATING PARAMETERS FOR GC/ECD ANALYSIS (METHOD FISHCF)

GC PARAMETER	SETTING	GC PARAMETER	SETTING
Temperature 1	80 degree C	Injector Temperature	220 degree C
Time 1	1 minute	Detector Temperature	325 degree C
Ramp 1	30 degree C per minute	Carrier Gas	Helium
Temperature 2	200 degree C	Constant Flow Pressure	A 20.1 psi /B 17.4 psi
Time 2	30 minutes	Constant Flow Rate	0.9 /0.7 mL per min
Ramp 2	2 degree C per minute	Septum Purge Flow Rate	4 mL per minute
Temperature 3	270 degree C	Make up Gas	Nitrogen
Time 3	0 minutes	Make up Gas Flow Rate	9.1/9.3 mL per minute
Ramp 3	10 degree C per minute	Injection Mode	Splitless for 1 minute
Temperature 4	280 degree C	Injection Volume	1.0 uL
Time 4	22 minutes		
Total run Time	93 min		
Constant Flow:	ON		

(NOTE: The operating parameters for both columns are the same).

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TABLE 2: GC AUTOSAMPLER OPERATING PARAMETERS FOR GC/ECD ANALYSIS

PARAMETER	SETTING	PARAMETER	SETTING
Inj/Bottle	1	Volume	1
First Bottle	1	# Of Solvent A washes	6 (Acetone)
Last Bottle	100	# Of solvent B washes	6 (Iso-Octane)
# Of Sample washes	2	Priority Sample (1=Yes)	No
# Of Pumps	6	Capillary On-Column (1=Yes)	No
Viscosity	1	Syringe	5 uL

TABLE 3: METHOD DETECTION LIMITS

	Dionex ASE 350 extraction				Soxhlet extraction			
ANALYTE	**MDL extract**	**PQL extract**	**MDL sample**	**PQL sample**	**MDL extract**	**PQL extract**	**MDL sample**	**PQL sample**
	(ug/L)	(ug/L)	(ug/kg)	(ug/kg)	(ug/L)	(ug/kg)	(ug/kg)	(ug/kg)
Aldrin	2.0	8.0	0.80	3.2	3.0	12	1.2	4.8
Alpha-BHC	2.5	10	1.0	4.0	2.5	10	1.0	4.0
Beta-BHC	3.0	12	1.2	4.8	7.0	28	2.8	11
Delta-BHC	4.0	16	1.6	6.4	5.0	20	2.0	8.0
Gamma-BHC	5.0	20	2.0	8.0	3.0	12	1.2	4.8
Cis-Chlordane	2.5	10	1.0	4.0	3.0	12	1.2	4.8
Cis-Nonachlor	5.0	20	2.0	8.0	6.0	24	2.4	9.6
DDD-o, p	5.0	20	2.0	8.0	5.0	20	2.0	8.0
DDD-p, p’	5.0	20	2.0	8.0	6.0	24	2.4	9.6
DDE-o, p	5.0	20	2.0	8.0	5.0	20	2.0	8.0
DDE-p, p’	5.0	20	2.0	8.0	6.0	24	2.4	9.6
DDT-o, p	5.0	20	2.0	8.0	8.0	32	3.2	13
DDT-p, p’	7.0	28	2.8	11	7.0	28	2.8	11
Dieldrin	2.5	10	1.0	4.0	5.0	20	2.0	8.0
Endosulfan I	2.0	8.0	0.80	3.2	2.5	10	1.0	4.0
Endosulfan II	2.0	8.0	0.80	3.2	2.5	10	1.0	4.0
Endosulfan Sulfate	6.0	24	2.4	9.6	9.0	36	3.6	14
Endrin	8.0	32	3.2	13	6.0	24	2.4	9.6
Heptachlor	3.0	12	1.2	4.8	3.0	12	1.2	4.8
Heptachlor Epoxide	2.5	10	1.0	4.0	2.0	8.0	0.80	3.2
Hexachlorobenzene	2.5	10	1.0	4.0	3.0	12	1.2	4.8
Mirex	7.0	28	2.8	11	8.0	32	3.2	13
Methoxychlor	15	60	6.0	24	22	88	8.8	35
Trans-Chlordane	2.5	10	1.0	4.0	4.0	16	1.6	6.4
Trans-Nonachlor	2.5	10	1.0	4.0	3.0	12	1.6	6.4
Toxaphene	75	300	30	120	75	300	30	120
PCB 8	4.0	16	1.6	6.4	5.0	20	2.0	8.0
PCB 18	3.0	12	1.2	4.8	4.0	16	1.6	6.4
PCB 28	4.0	16	1.6	6.4	5.0	20	2.0	8.0
PCB 52	2.5	10	1.0	4.0	5.0	20	2.0	8.0
PCB 44	2.5	10	1.0	4.0	4.0	16	1.6	6.4
PCB 66	2.5	10	1.0	4.0	4.0	16	1.6	6.4
PCB 77	2.5	10	1.0	4.0	4.0	16	1.6	6.4

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PCB 101	4.0	16	1.6	6.4	4.0	16	1.6	6.4
PCB 118	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 126	2.5	10	1.0	4.0	4.0	16	1.6	6.4
PCB 153	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 105	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 138	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 187	3.0	12	1.2	4.8	4.0	16	1.6	6.4
PCB 128	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 180	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 170	2.5	10	1.0	4.0	3.0	12	1.2	4.8
PCB 195	3.0	12	1.2	4.8	3.0	12	1.2	4.8
PCB 206	2.5	10	1.0	4.0	3.0	12	1.2	4.8

TABLE 4: STANDARD CONCENTRATION LEVELS

ANALYTE	**STANDARD SOLUTIONS (ug/L)**								**SPK SOLUTION (ug/L)**
ECD A, C and F MIXTURES	Inter-mediate	Dil. 1	Dil 2	Dil 3	Dil 4	Dil 5	Dil 6	Dil 7	
Aldrin	1,000	50	37.5	25	10	4	2		100
Alpha-BHC	1,000	50	37.5	25	10	4	2		100
Beta-BHC	1,000	50	37.5	25	10	4	2		100
Delta-BHC	1,000	50	37.5	25	10	4	2		100
Gamma-BHC	1,000	50	37.5	25	10	4	2		100
Cis-Chlordane	1,000	50	37.5	25	10	4	2		100
Cis-Nonachlor	2,000	100	75	50	20	8	4		200
DDD-o, p	2,000	100	75	50	20	8	4		200
DDD-p, p’	2,000	100	75	50	20	8	4		200
DDE-o, p	2,000	100	75	50	20	8	4		200
DDE-p, p’	2,000	100	75	50	20	8	4		200
DDT-p, p’	2,500	100	75	50	20	10	5		200
DDT-o,p	2,000	100	75	50	20	8	4		
Dieldrin	1,000	50	37.5	25	10	4	2		100
Endosulfan I	1,000	50	37.5	25	10	4	2		100
Endosulfan II	1,000	50	37.5	25	10	4	2		100
Endosulfan Sulfate	2,000	100	75	50	20	8	4		200
Endrin	2.500	100	75	50	20	10	5		200
Heptachlor	1,000	50	37.5	25	10	4	2		100
Heptachlor Epoxide	1,000	50	37.5	25	10	4	2		100
Hexachlorobenzene	1,000	50	37.5	25	10	4	2		100

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ANALYTE	STANDARD SOLUTIONS (ug/L)								SPK SOLUTION (ug/L)
Mirex	2,000	100	75	50	20	10	5		200
Methoxychlor	5,000	250	187.5	120	50	20	10		500
Trans-Chlordane	1,000	50	37.5	25	10	4	2		100
Trans-Nonachlor	1,000	50	37.5	25	10	4	2		100
Toxaphene	10,000	5,000	2,000	1,000	600	300	200		10,000
CONGENERS MIXTURE									
PCB 8	1,000	100	75	50	25	10	5	5	100
PCB 18	1,000	100	75	50	25	10	5	5	100
PCB 28	1,000	100	75	50	25	10	5	5	100
PCB 52	1,000	100	75	50	25	10	5	5	100
PCB 44	1,000	100	75	50	25	10	5	5	100
PCB 66	1,000	100	75	50	25	10	5	5	100
PCB 77	1,000	100	75	50	25	10	5	5	100
PCB 101	1,000	100	75	50	25	10	5	5	100
PCB 118	1,000	100	75	50	25	10	5	5	100
PCB 153	1,000	100	75	50	25	10	5	5	100
PCB 105	1,000	100	75	50	25	10	5	5	100
PCB 138	1,000	100	75	50	25	10	5	5	100
PCB 187	1,000	100	75	50	25	10	5	5	100
PCB 128	1,000	100	75	50	25	10	5	5	100
PCB 126	1,000	100	75	50	25	10	5	5	100
PCB 180	1,000	100	75	50	25	10	5	5	100
PCB 170	1,000	100	75	50	25	10	5	5	100
PCB 195	1,000	100	75	50	25	10	5	5	100
PCB 206	1,000	100	75	50	25	10	5	5	100
PCB 209	1,000	100	75	50	25	10	5	5	100

TABLE 5: RESISTANCE TO SULFURIC ACID CLEAN UP

Parameter	%Recovery after Sulfuric acid	Parameter	%Recovery after Sulfuric acid
Aldrin	Non-resistant	Endrin	Non-resistant
Alpha-BHC	Resistant	Chlordane	Resistant
Beta-BHC	Resistant	PCB-1016	Resistant
Delta-BHC	Resistant	PCB-1221	Resistant

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Gamma-BHC	Resistant	PCB-1232	Resistant
DDD-p, p’	Resistant	PCB-1242	Resistant
DDD-o, p’	Resistant	PCB-1248	Resistant
DDE-p, p’	Resistant	PCB-1254	Resistant
DDE-o, p’	Resistant	PCB-1260	Resistant
DDT-p, p’	Resistant	Toxaphene	Resistant
DDT-o, p’	Resistant	Cis-Chlordane	Resistant
Dieldrin	Non-resistant	Trans-Chlordane	Resistant
Endosulfan I	Non-resistant	Cis-Nonachlor	Resistant
Endosulfan II	Non-resistant	Trans-Nonachlor	Resistant
Endosulfan Sulfate	Non-resistant		
Endrin Aldehyde	Non-resistant	Hexachlorobenzene	Resistant
Heptachlor	Resistant	Gamma-Chlordane	Resistant
Heptachlor Epoxide	Non-resistant	Mirex	Resistant
Endrin Ketone	Non-resistant	Methoxychlor	Non-resistant

TABLE 7: QC MEASUREMENTS, ACCEPTANCE CRITERIA AND CORRECTIVE ACTIONS

#	QC MEASUREMENT	ACCEPTANCE CRITERION	CORRECTIVE ACTION
1	LFB/Matrix Spike* - Recoveries - Precision Endrin Methoxychlor	60-130% =/< 30% 70-150% 70-145%	Reanalyze/Re-extract/Flag
2	CCV	+/- 15%	Re-run and recalibrate/Flag
3	QCS verification	+/- 15%	Re-run and recalibrate/Flag
4	Surrogate recovery DCB TMX	 76-146% 56-115%	Reanalyze/Re-extract/Flag
5	Linearity criterion Non-Linear	20% RSD 0.995 CC	Use appropriate cal fit / Re-run/ Recalibrate
6	DDT/Endrin %Dec	15 % each	Maintain /Rerun/Recalibrate/Flag

- * Statutory limits are used for assessing LFB/matrix spike recoveries. The limits are verified annually based on statistical data. Statistical limits are used whenever they differ significantly from the statutory limits.

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APPENDIX OF CHANGES

09/15/2010	Added Appendix 1 Standard Preparation Procedure.
01/28/2011	Reference to check the calibration standards vs. QCS once per month have been removed. QC Manager Instrument "GCPesticides:Target" has been changed to "GC_ECD_5 (or other GC_ECD_#). Surrogates Recoveries have been updated.
10/08/2012	MDLs for Dionex ASE 350 extractions added. Cleanup procedures updated.
09/30/2013	Requirement for six levels for quadratic calibration have been added. The run sequence have been rearranged to put all CCVs just before the samples. The GC program have been updated. The statistical limits formula for surrogate recoveries have been updated.

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