Extraction, Clean-Up And Analysis Of All 209 PCBs With Simultaneous Separation Of PCDD/Fs In One Run Using LCTech DEXTech Systems

Dr. Thomas Fiedler, Thomas Kerkemeier, Dr. Uwe Aulwurm

Content

Introduction

The analysis of dioxins (polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) as well as all 209 polychlorinated biphenyls (PCBs) in environmental and food samples is paramount in safeguarding human health and environmental integrity.

Dioxins and PCBs are classified as persistent organic pollutants (POPs), characterized by their resistance to degradation and ability to bioaccumulate in the environment, posing significant risks to both ecosystems and human health. In light of their widespread distribution and harmful effects, comprehensive testing for these contaminants is essential across various environmental and food matrices.

Environmental samples, including soil, biota, sediments, and air, serve as crucial indicators of the presence and distribution of dioxins and PCBs in ecosystems. These pollutants can originate from various sources, including industrial activities, waste incineration, and unintentional byproducts of chemical processes. Once released into the environment, dioxins and PCBs can undergo long-range transport, contaminating remote regions far from their original sources. Therefore, testing environmental samples enables the identification of contamination hotspots, assessment of pollutant dispersion patterns, and evaluation of potential ecological risks.

In particular, soil samples and biota play a vital role in understanding the fate and behavior of dioxins and PCBs in terrestrial ecosystems. Due to their high affinity for organic matter and soil particles, these contaminants can persist in soil for extended periods, posing risks to soil-dwelling organisms and potentially entering the food chain. By testing soil samples, researchers and environmental authorities can monitor contamination levels, track pollutant migration pathways, and inform remediation efforts aimed at mitigating environmental impacts.

In addition to environmental samples, testing dioxins and PCBs in food samples is essential for ensuring food safety and protecting human health. These contaminants can accumulate in the fatty tissues of animals and bioaccumulate in the food chain, resulting in potentially hazardous levels in foods such as meat, dairy products, fish, and other seafood. Chronic exposure to dioxins and PCBs through contaminated food consumption has been linked to adverse health effects, including developmental abnormalities, reproductive disorders, immune system impairment, and cancer.

Therefore, rigorous testing of food samples is critical for identifying contaminated food products, assessing human exposure risks, and enforcing regulatory limits to protect public health. By analyzing dioxins and all 209 PCB congeners in food matrices, regulatory authorities can ensure compliance with safety standards, implement risk management measures, and safeguard consumer health.

Through coordinated efforts in environmental monitoring, food safety regulation, and risk assessment, stakeholders can work towards minimizing the adverse impacts of these persistent pollutants on ecosystems and human populations.

The U.S. Environmental Protection Agency (EPA) has developed several methods for testing dioxins and PCBs in environmental and food samples. Some commonly used methods include:

- EPA Method 1613B: This method is used for the analysis of dioxins and furans in environmental samples such as soil, sediment, and tissue. It involves sample extraction, clean-up, and analysis using high-resolution gas chromatography/mass spectrometry (HRGC/HRMS).
- EPA Method 1668C: This method is specifically designed for the analysis of PCB congeners in environmental samples such as water, sediment, soil, and tissue. It utilizes high-resolution gas chromatography/mass spectrometry (HRGC/HRMS) to separate and quantify individual PCB congeners.
- EPA Method 8290A: This method is used for the analysis of PCDD/Fs in solid waste samples, such as incinerator ash and soil. It employs HRGC/HRMS for the separation and quantification of target compounds.

These EPA methods provide standardized procedures for the accurate and reliable analysis of dioxins and PCBs in various environmental matrices.
Automated sample preparation equipment can save precious time and resources in daily operation. They help to standardize method and also help to lower manual handling errors. Additionally, today's laboratories require equipment, where global regulations and changing methods can be implemented easily into their own processes.
In this application note we will demonstrate the extraction, evaporation, clean-up and analysis of all 209 PCBs and PCDD/F from different matrices like fish oil and soil using LCTech unique automated sample prep instruments. (see Figure 1).

Figure 1. The automated workflow for the extraction, evaporation and clean-up of a variety of POPs (PCBs, PCDD/Fs, PBDEs and PCNs) using LCTech instruments

Materials and Methods

Reagents and Materials

- Standard solutions
  - EPA1613-PAR/Stock, Wellington Laboratories
  - PCB-LCS-H and PCB-ISS-H Wellington Laboratories
  - PCB-Stock-A20, Wellington Laboratories
  - EDF-5526-100x, Recovery Standard, CIL
  - EDF-5525-100x Internal Standard, CIL

- Solvents
  - Hexane (e.g. VWR – picograde, or neofroxx hplcgrade)
  - Toluene (e.g. VWR – picograde or supelco suprapur)
  - Dichloromethane (e.g. VWR – picograde or supelco suprapur)
  - Dodecane (optional as keeper)

- X-TRACTION®, LCTech GmbH
  - Extraction cell, 75 mL (nominal volume) p/n 19700
  - Glass fiber filter (37 mm diameter) p/n 19281
  - Stainless steel Frits 10 µm p/n 17782

- DEXTech Pure & Heat, LCTech GmbH
- 3 Columns set-up for clean-up
  - 1st column (209PCB p/n 20325 - LCTech GmbH)
  - 2nd column (alumina p/n 15433 - LCTech GmbH)
  - 3rd column (carbon p/n 21344 - LCTech GmbH)

- D-EVA, powered by LCTech GmbH
  - Rotational-Vacuum-Concentrator D-EVAporation, complete and ready to use, consists of concentrator with special software for dioxin and PCB evaporation, cooling trap, pressure control, vacuum pump (p/n 16900 LCTech)
  - Rotor (p/n 16929)
    - Centrifuge vials (130 mL p/n 16725 LCTech)
    - Temperature sensor (p/n 16755 LCTech)
  - Rotor (p/n 16742)
    - Centrifuge vials (15 mL, p/n 15781 LCTech)
    - Temperature sensor (p/n 16741 LCTech)

- Rotors (p/n 16802)
  - Centrifuge vials (40 mL p/n 16452 LCTech)
  - Temperature sensor (p/n 16738 LCTech)

- DFS HRMS, Thermo Fisher Scientific
  - SSL, HT8-PCB, 60 m, 0.25 mm film, 25 mm ID, Trajan
  - PTV, RTX-Dioxin2, 60 m, 0.25 mm film, 25 mm ID, Restek

Extraction

This chapter describes how PCBs and PCDD/Fs were extracted of soil samples using the X-TRACTION® system from LCTech (Figure 2). This system relies on low-pressure liquid extraction (LPLE) to remove PCBs and PCDD/Fs from the sample. To begin, 10 grams of dried soil were sieved and combined with 5 grams of sodium polyacrylate. This mixture was then loaded into the extraction cell (see Figure 3) and sealed shut using the specialized magnetic closure of the LCTech extraction cell.

Figure 2 shows the basis unit of the X-TRACTION® system from LCTech

Figure 3 shows the extraction with holder and the unique magnetic lid for the easy closure of the cell

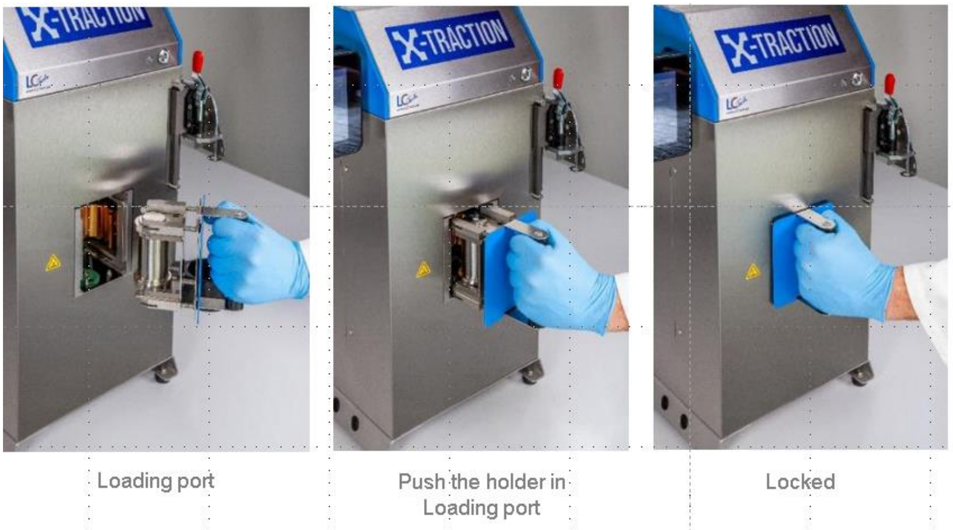


Figure 4 shows the insertion of the extraction cell into the X-TRACTION® system.

Next, the cell was inserted into the X-TRACTION® system (refer to Figure 4), and the soil underwent extraction with toluene, following the parameters outlined in Figure 5.

The extraction process involved two cycles. In each cycle, the extraction cell received first 20 mL of toluene from the top and than from the bottom. Heating the cell to 150°C for 7 minutes facilitated the extraction, followed by flushing with an additional 10 mL of toluene and a two minute nitrogen blowout. Overall, the extraction of one soil sample took approximately 32 minutes. Post-extraction, the soil samples (approximately 80 mL) were evaporated and concentrated to a smaller volume using the LCTech D-EVA system.

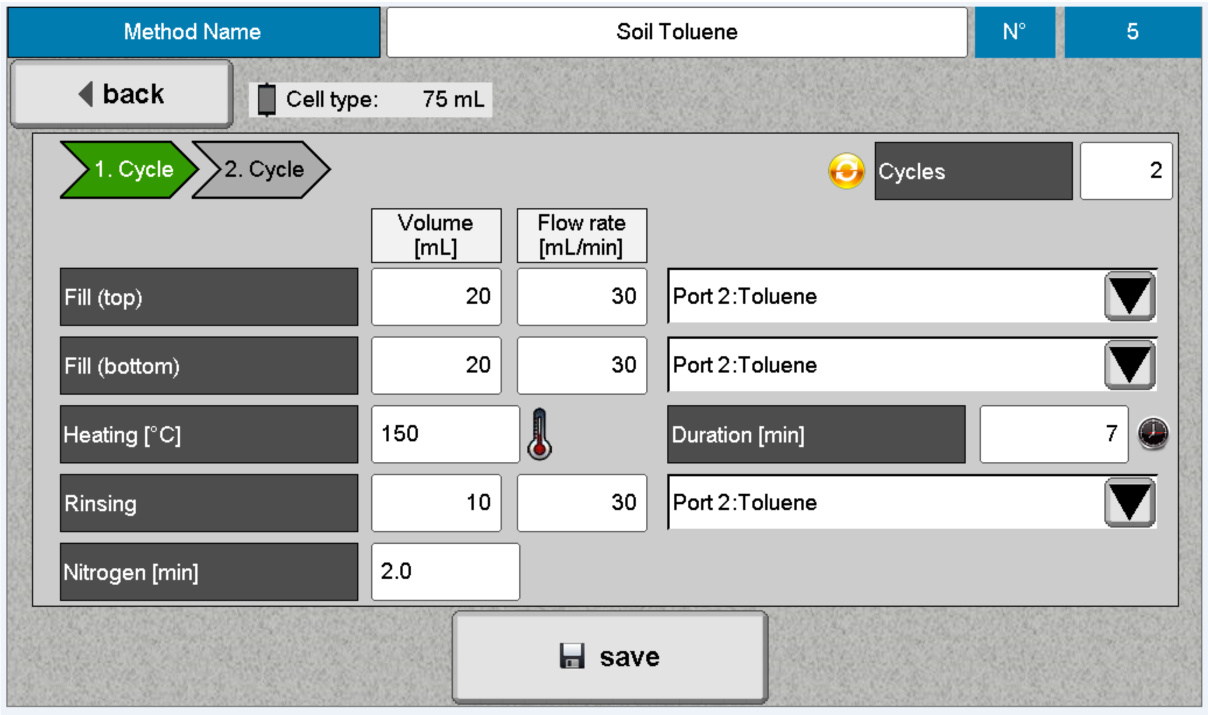


Figure 5 shows the process window in the X-TRACTION® software with the used parameters for the soil extraction.

In contrast, the fish oil samples did not undergo extraction. Instead, 3 grams of fish oil were mixed with 1 mL of toluene and topped up with n-hexane to a total volume of 15 mL. This prepared sample was then directly used for the cleanup procedures.

(if you would like to know more about the X-TRACTION system please go to our website under : Link)

Evaporation

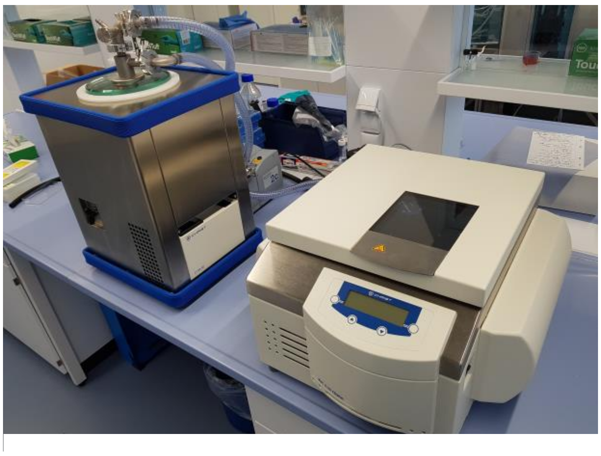


Figure 6 shows the D-EVA with cryotrap and vacuum pump

If an extraction of the sample is needed, there are 2 evaporation steps during the whole workflow. The first after the extraction, the second after the clean-up.

Evaporation after Extraction:
The evaporation after the extraction is a solvent exchange to n-hexane. 90 mL of toluene and matrix must be evaporated to a final volume of 300-500 µL. A few drops of dodecane as keeper has been added before the evaporation. Using the

D-EVA with the program in Table 1 the final volume of the evaporation has been 800 - 1500 µL. By using a nitrogen blow down, the final volume of less than 500 µL has been achieved. Subsequently, the sample has been transferred to the sample vial of the DEXTech heat followed by three times of rinsing the centrifuge tube with 2 – 3 mL n-hexane for each transfer. The sample has been filled up to 15mL for the clean-up, that is described in the next chapter.

Evaporation after Clean-up:
After the clean-up there are two fractions. The first fraction 1 (F1) with all 209 PCB in 24 mL toluene has to be evaporated down to 100 µL. The final volume of the second fraction 2 (F2) with PCDD/F in 11 mL toluene should be at 20 µL. To avoid losses of the highly volatile mono- and di-chlorinated PCBs, it is important to evaporate gently and not to complete dryness. Both fractions were spiked with recovery standard solutions right before the D-EVA. This was done in order to make a more accurate judgement about the efficiency of the DEXTech System and the X-TRACTION®.

The evaporation of fraction 1, containing the PCB, has been stopped at 100 – 200 µL, for better $^{13}C_{12}$ recoveries of the lower chlorinated PCB and blown down to 100 µL with a gentle stream of nitrogen. Fraction 2 has been evaporated down to near dryness directly via D-EVA, and transferred to the GC-vials.

The following toluene program has been used for all evaporations.

Section	Unit	Start	1	2	3
Time	h:m		00:03	00:10	02:00
Temperature	°C	40	40	40	40
Pressure	mbar		30	10	10
Sa.pressure	mbar		200	80	500
Speed	min^{-1}		800	800	800

Stop at 30°C

Table 1 shows the D-EVA program used for the sample evaporation

Sample Clean-Up

Fish oil samples were prepared by measuring 3 g of fish oil, adding 1 mL of toluene, and then filling the mixture up to a final volume of 15 mL with n-hexane. Prior to the clean-up, the fish oil samples were spiked with $^{13}C_{12}$ -PCB and $^{13}C_{12}$ -PCDD/F standards, along with native PCBs and PCDD/Fs. Similarly, the soil samples underwent the same spiking process before extraction.

Figure 9 : DEXTech Heat system

For the clean-up process, the samples were placed into the DEXTech Heat system (refer to Figure 9) along with three clean-up columns. The first column was an acidic silica column (PCB 209 column p/n 20325), the second was an alumina column (p/n 15433), and the third column was a carbon column (p/n 21344). The PCB209+Dioxin method was selected for the clean-up process (refer to Figure 10 a and b).

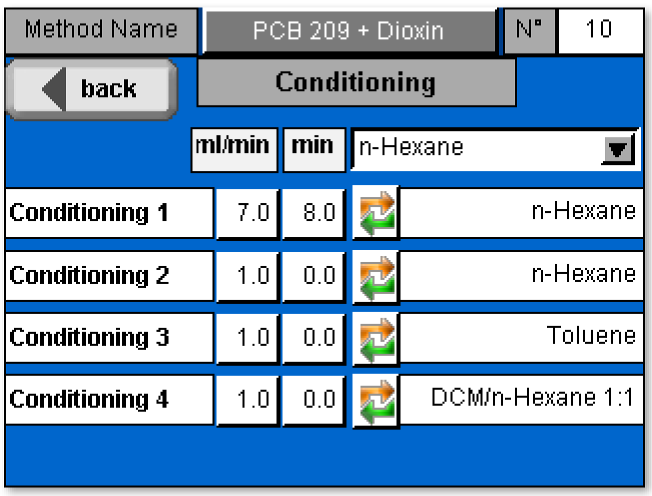


Figure 10 a Conditioning window

Figures 10 a and 10 b illustrate the processing window of the PCB 209 method in the DEXTech Heat software. A schematic description of the flow path for the method is available in the supplementary materials.

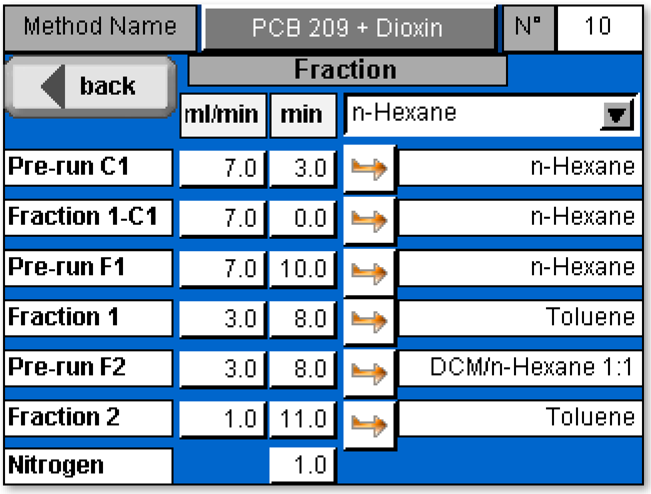


Figure 10 b. Fractionation window of the PCB 209 +Dioxin method

During this method, the samples are not only cleaned for analysis, but also all 209 PCBs are separated from the PCDD/Fs. This method yields two fractions: one fraction containing all the 209 PCBs (24 mL) and a second fraction containing the PCDD/Fs (11 mL). The method commences with conditioning column 1 with n-hexane (see Figure 10 a). After the conditioning step, the sample is loaded from the sample vial over the sample loop onto column 1 (PreRun C1). During the loading process, the sample vial is automatically rinsed 3 time with 1 mL of n-hexane. The wash solution is added to the sample in the sample loop and transferred onto column 1. This ensures that more than 99% of the sample is transferred onto the column 1.

Subsequently, the PCBs and PCDD/Fs are flushed from the acidic silica column onto the alumina column (column 2) (PreRunF1). Following this, all 209 PCBs are eluted in fraction 1 with toluene from the alumina column, while the PCDD/Fs remain retained on the alumina column. The PCDD/Fs are then flushed from the alumina column with a mixture of dichloromethane (DCM) and n-hexane (50:50%) onto the carbon column and subsequently eluted in fraction 2 with toluene. The processing time for this method is approximately 52 minutes. If only fraction 1 containing all the 209 PCBs is required for analysis, only columns 1 and 2 are utilized, reducing the processing time to 33 minutes. For analytical purposes, the two fractions need to be concentrated by evaporating them down using the LCTech D-EVA system (see Evaporation section).

Instrumental Analysis

The analysis of PCDD/F and all 209 PCB was carried out using the DFSTM Magnetic Sector GC-HRMS system from Thermo Fisher Scientific. The DFS is equipped with 2 Trace 1310 gas chromatographs. For the measurement of PCDD/F, the Rtx - Dioxin2 from Restek via PTV and the 209 PCB a SSL injector with the SGE HT8-PCB from Trajan was used. Both columns are 60 m columns with an inner diameter of 0.25 mm and a film thickness of 0.25 µm. The injection volume for the PCDD/F is 5 µL and for the PCB 2 µL. More details like GC oven condition, injector program, autosampler parameter and MID are to be found in the Appendix. The elution orders of the PCDD/F and all 209 PCB can be found in the appendix as well.

Quality Check:
Based on the signal/noise ratio the LOD has been calculated with TargetQuan, this software is provided and validated by Thermo Fisher Scientific. The isotope ratio is checked for every signal by TargetQuan as well as the relative retention time. The response factors (RF) are determined every day with a midrange calibration standard solution called verification check (VER) and recorded in a control chart. Especially for the analytic of the 209 PCB the peak shape and resolution power of the GC-column are essential. With the SGE HT8-PCB from Trajan the #28 and #31 have to be baseline separated. The Rtx Dioxin2 from Restek should have a maximum overlay of the 1,2,3,4,7,8-HxCDF and 1,2,3,6,7,8-HxCDF of 10 %.

Response factors

As part of the Quality Check response factors are calculated by measuring a VER-Check. This has been done for every native and for any $^{13}C_{12}$ -labelled compound.

$$\mathrm{RF} = \frac{\mathrm{AA_{LCS}} \times \mathrm{c_{ISS}}}{\mathrm{AA_{ISS}} \times \mathrm{c_{LCS}}}$$

AA_{ISS} areas of Internal Standards (recovery)
AA_{LCS} areas Labelled Compounds (quantifier)
c_{ISS} certified concentration of Internal Standards
c_{LCS} certified concentration of Labelled Compounds

$^{13}C_{12}$ Recoveries

As the labelled compound solution LCS with known concentration is added to the sample right after the sample intake, all labelled substances undergo the same process as the origin native analytes. Any influences of the process affect these labelled compounds the same way as the analytes. By adding the internal standard solution right at the end of the process as 100 % standard these influences of the process can be quantified. The concentration of the labelled compound was calculated by following formula and compared with the theoretical amount of the analyte.

$$c_L = \frac{\mathrm{AA_{LCS}} \times \mathrm{c_{ISS}}}{\mathrm{AA_{ISS}} \times \mathrm{RF}} \qquad \frac{c_L}{c_{LCS}} \times 100 = Recovery\ [\%]$$

AA_{ISS} areas of Internal Standards (recovery)
AA_{LCS} areas Labelled Compounds (quantifier)

RF	response factors from the VER Check
c_{ISS}	certified concentration of Internal Standards
c_{LCS}	certified concentration of Labelled Compounds
c_L	calculated concentration of Labelled Compound

Native Results

The native concentration in the sample can be calculated by following formula:

$$\mathrm{RF} = \frac{\mathrm{AA_n} \times \mathrm{c_{LCS}}}{\mathrm{AA_{LCS}} \times \mathrm{c_n}} \qquad c = \frac{\mathrm{AA_n} \times \mathrm{c_{LCS}}}{\mathrm{AA_{LCS}} \times \mathrm{RF}}$$

AA_{LCS}	areas Labelled Compounds (quantifier)
AA_n	areas of Native Analytes
c_{LCS}	certified concentration of Labelled Compounds
c	calculated concentration of native Compounds

Results

The aim of this study was to develop an automated clean-up method for all 209 PCBs in accordance with quality acceptance criteria (Table 6) of EPA method 1668C, and for the simultaneous clean-up and separation of PCDD/Fs. While the EPA methods describe the cleanup and analysis of either all 209 PCBs or PCDD/Fs separately, this automated method is designed for the simultaneous clean-up and separation of both PCBs and PCDD/Fs. This has the potential to increase production efficiency, sample output and thus reduce costs in the lab.

A total of 5 blank samples, 2 different soil samples (each n=6), and 1 fish oil sample (n=9) were evaluated for initial recovery and precision (IRP) of all native 209 PCBs and 17 PCDD/Fs, as well as $^{13}C_{12}$ labeled PCBs and PCDD/Fs.

Initial Recovery and Precision of $^{13}C_{12}$ -Labeled Compounds in Blank, Soil and Fish Oil Samples

The quality control (QC) acceptance criteria for initial recovery and precision (IRP) of labeled $^{13}C_{12}$ -PCB standards and native PCBs are outlined in EPA method 1668C (Table 6). The mean recovery for labeled standards should fall within the range of 45 % to 135 %, while for native PCBs, it should be between 70 % and 130 %. The relative standard deviation (RSD) (%) values for labeled compounds should be below 50 %, or 25 % for native PCBs.

In accordance with EPA method 8290A, a recovery range of 40 % to 135 % is required for all 2,3,7,8-substituted internal $^{13}C_{12}$ -standards. Similarly, EPA method 1613B specifies a recovery range of 72 % to 152 %, depending on the native PCDD/F congener (refer to supplementary materials for details).

Figures 11 and 12 illustrate a graphical summary of the mean recovery rates for the $^{13}C_{12}$ -labeled PCDD/F (Figure 11) and PCB (Figure 12) congeners used in the study. Error bars indicate the relative standard deviation (RSD) (%) values for the different congeners in the different matrices.

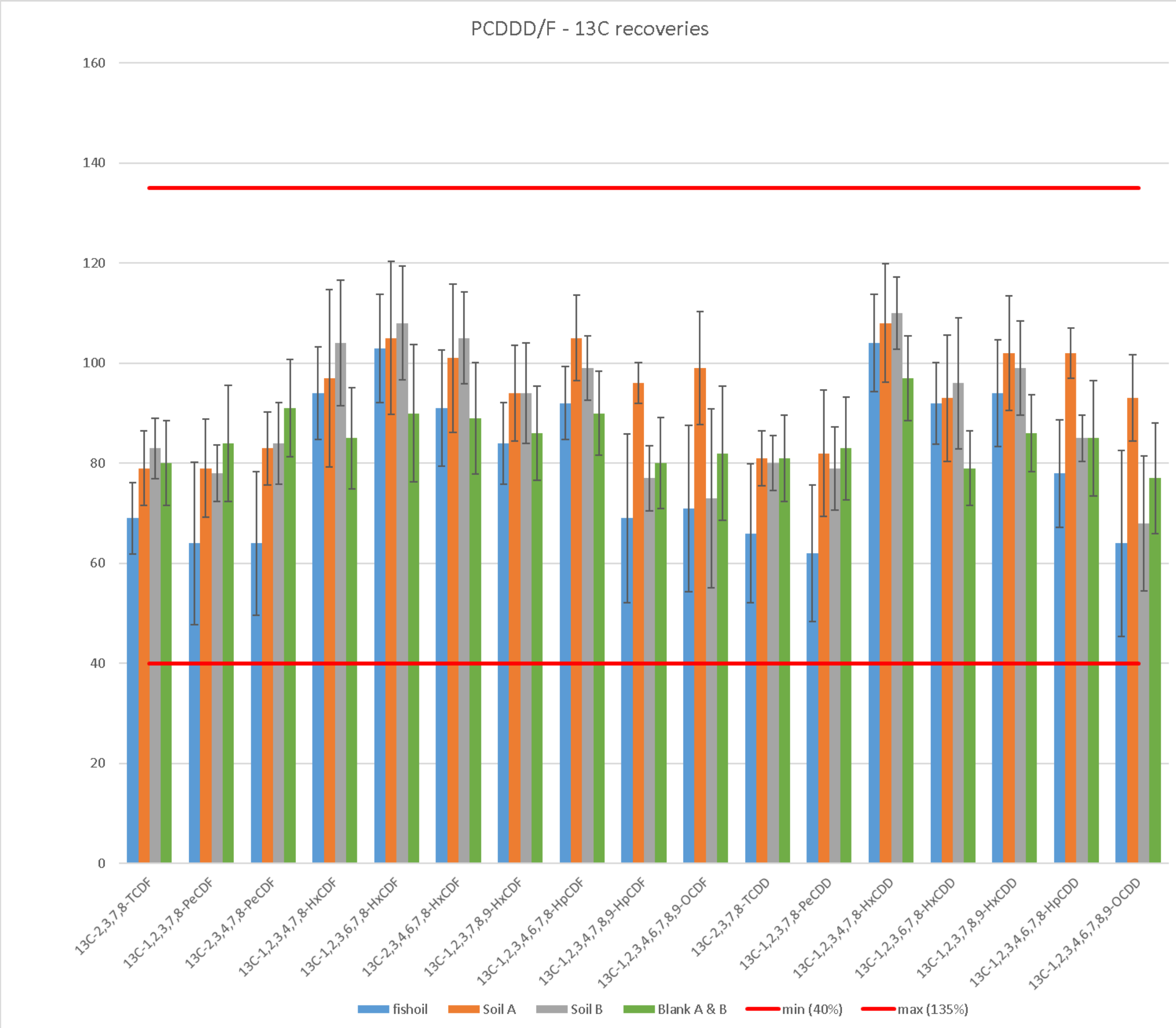


Figure 11 ^{13}C-PCDD/F recoveries for blanks, fish oil and 2 different soil samples. (min bar = 40%, max bar = 135 %)

For soil sample A (n=6), the mean recoveries of the different PCDD/Fs congeners ranged from 79 % ($^{13}C_{12}$ -2,3,7,8-TCDF and $^{13}C_{12}$ -1,2,3,7,8-PeCDF) to 108 % ($^{13}C_{12}$ -1,2,3,4,7,8-HxCDD). Soil sample B (n = 11) exhibited recoveries of 68 % for $^{13}C_{12}$ -1,2,3,4,6,7,8,9-OCDD and 110 % for $^{13}C_{12}$ --1,2,3,4,7,8-HxCDD. The fish oil recoveries (n = 9) ranged from 62 % for the $^{13}C_{12}$ -1,2,3,7,8-PeCDD congener to 104 % for the ^{13}C-1,2,3,4,7,8-HxCDD. RSD (%) values varied among the samples, with the highest RSD of 18.6 % for the ^{13}C-1,2,3,4,6,7,8,9-OCDD in the fish oil sample and 4.1 % for the ^{13}C-1,2,3,4,7,8,9-HpCDF congener in soil sample A. In summary, the RSD values and recoveries were compliant with the quality control (QC) criteria for labeled standards outlined in EPA methods 1613 and 8290A.

Figure 12 presents the summarized results of the labeled $^{13}C_{12}$ -PCB recoveries. All $^{13}C_{12}$ -labeled PCBs exhibited recoveries above 45 % and below 135 %. The only exception was the mean recovery of $^{13}C_{12}$

-labeled PCB 4 in soil B, which was below the 45 % threshold at 40 %. This discrepancy likely stems from a suppression due to a contamination originating in the laboratory, as purity and efficiency checks of the clean-up columns conducted by a collaborating laboratory did not indicate such suppression. However, despite this suppression, the recoveries still met the QC criteria for $^{13}C_{12}$ -labeled PCBs outlined in EPA method 1668C (Table 6 #PCB 4 20-135%). Even with this anomaly, the RSD values for all $^{13}C_{12}$ -labeled PCBs remained below 50 %, ranging from 1.8 % to 27.9 %, thereby satisfying the acceptance criteria for $^{13}C_{12}$ -labeled PCBs in the US EPA Method 1668C.

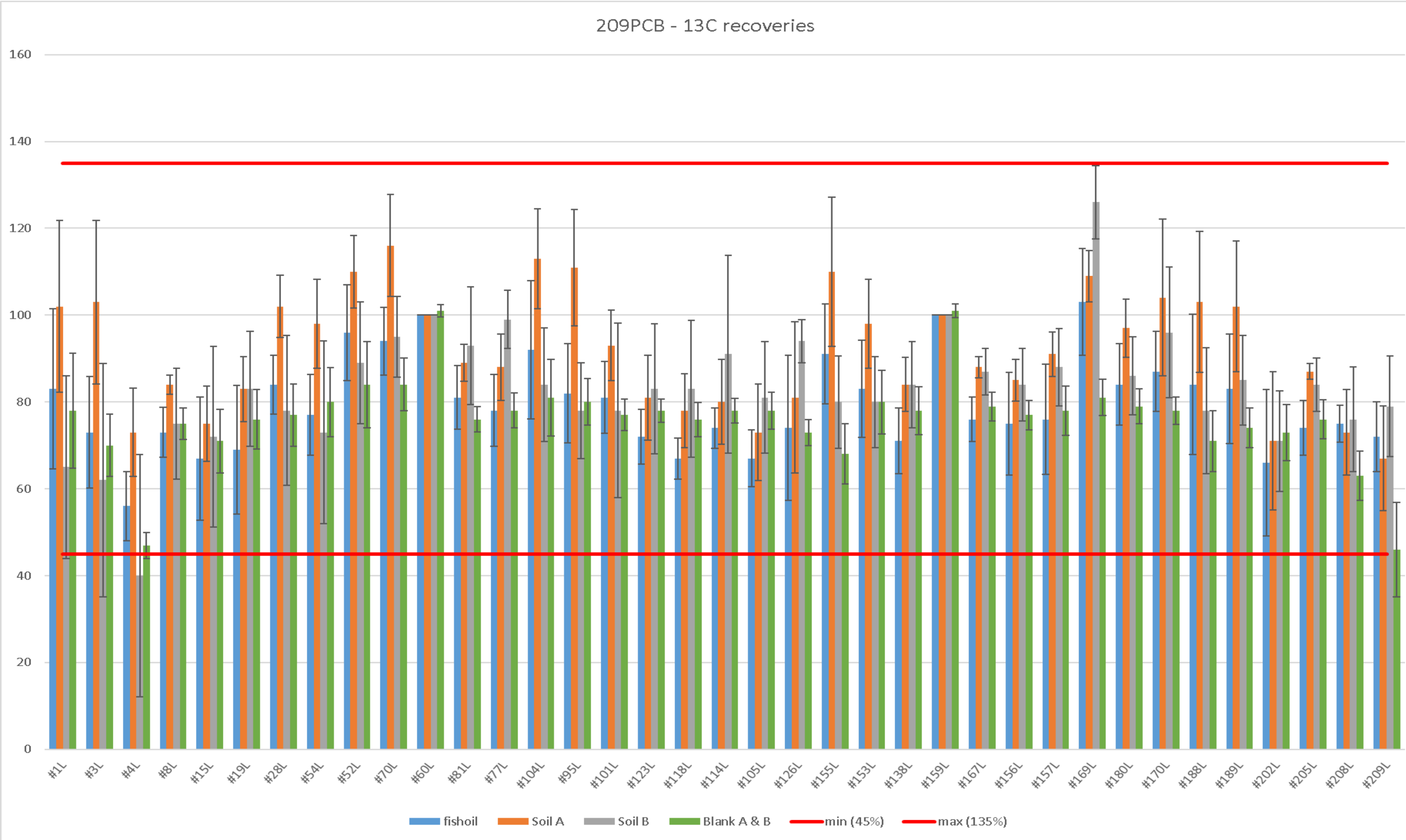


Figure 12 PCBs $^{13}C_{12}$ recoveries

Initial Recovery and Precision of Native Fortified PCBs and PCDD/Fs in Blank, Soil and Fish Oil Samples

In the initial fortification experiments, we conducted triplicate measurements of soil samples A and B, as well as the fish oil sample, to establish the background levels of PCBs and PCDD/Fs in each sample. Subsequently, each sample was fortified with 4000 pg absolute of all 209 native PCBs. The fortification level of the native TCDD/Fs is 750 pg absolute, the native PnCDD/F to HpCDD/F 3500 pg absolute and the native OCDD/F with 6500 pg absolute. For fish oil, n = 4; for soil A, n = 4; for soil B, n = 11 were tested.

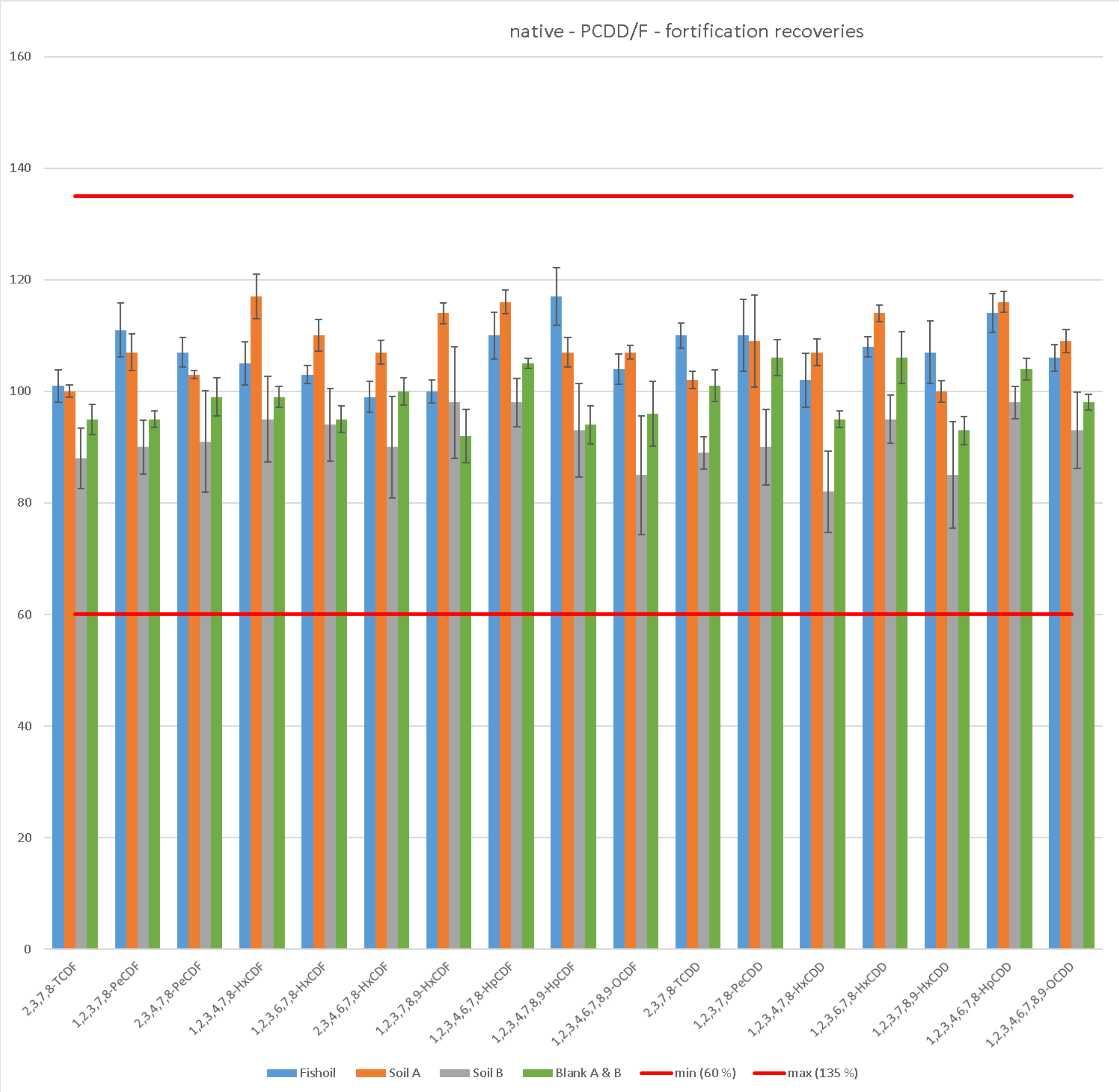


Figure 13 native PCDDF/Fs recoveries of the different fortified samples

Figure 13 provides a summary of the mean recovery rates for the fortified native PCDD/Fs across the different matrices.

According to EPA 1613B, the acceptance criteria for recoveries of native PCDD/Fs in fortified samples vary depending on the congener, ranging between 72 % and 150 % (see supplementary material). The recoveries for all PCDD/F congeners in the different matrices fell within the range of 82 % to 117 %, with RSD values ranging from 0.7 % to 10.6 %, thereby meeting the acceptance criteria specified in EPA method 1613B.

In EPA method 1668C, Table 6 outlines the acceptance criteria for native PCBs in the initial recovery and precision (IPR) experiments. The mean recoveries should fall within the range of 70 - 130 %, and the relative standard deviation (RSD) value should be below 25 %. Figure 14 presents the results of the recoveries of native PCBs in our tests. The exact values for each PCB are also provided in Table X of the supplementary materials. As shown in the graph, the recoveries of all native PCBs were above 70 % and below 130 %, with the lowest recovery observed for PCB 149/139 in fish oil (71 %) and the highest recovery for PCB 50 in fish oil (130 %). The RSD values for all the PCBs were below 25 %, ranging from 0.8 % to 24.1 %. In summary the results are in accordance with the quality control criteria of EPA 1668C (Table 6).

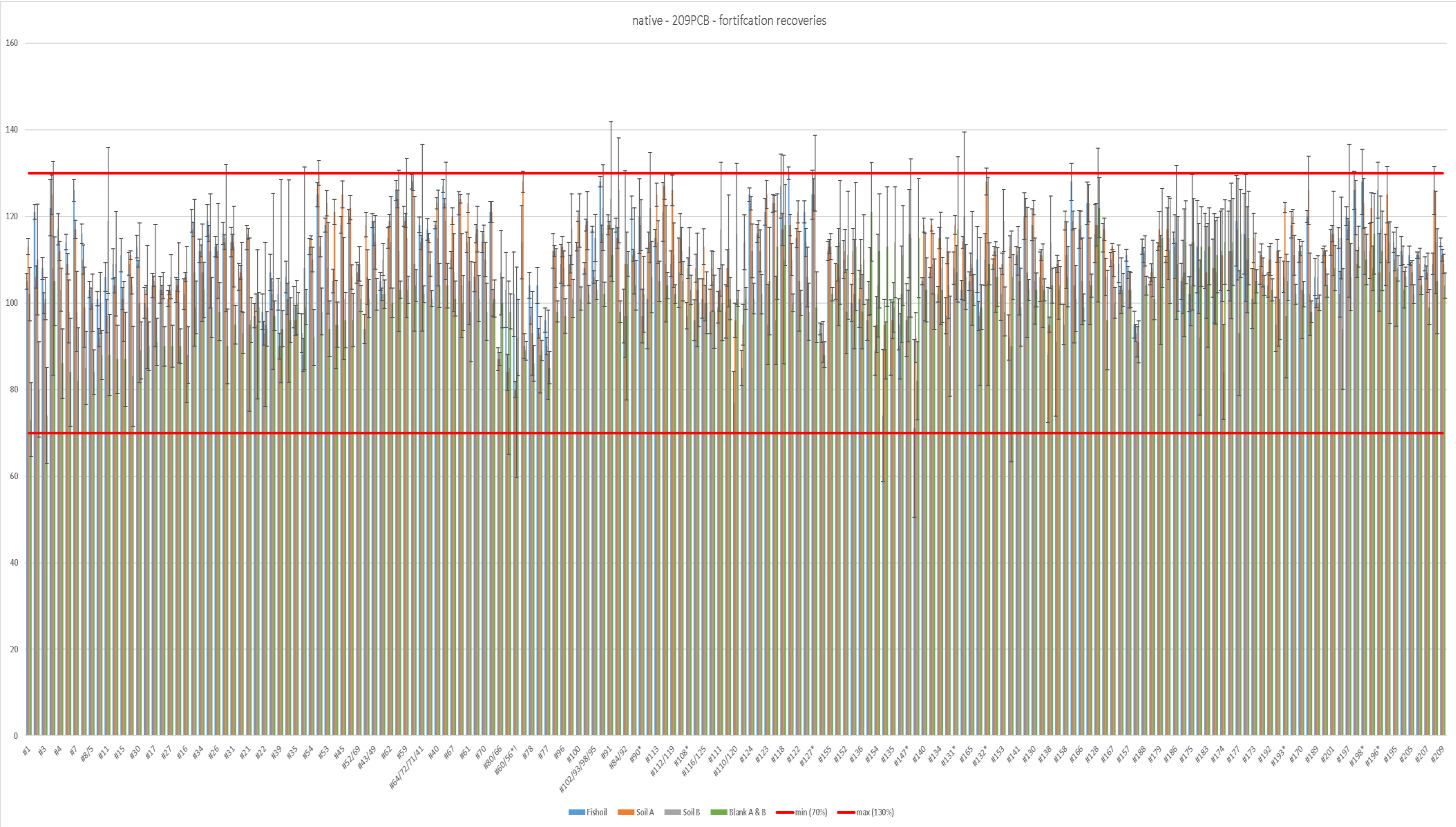


Figure 14 graphical representation of the mean recoveries for all 209 PCBs in the different sample.

Summary and Conclusion

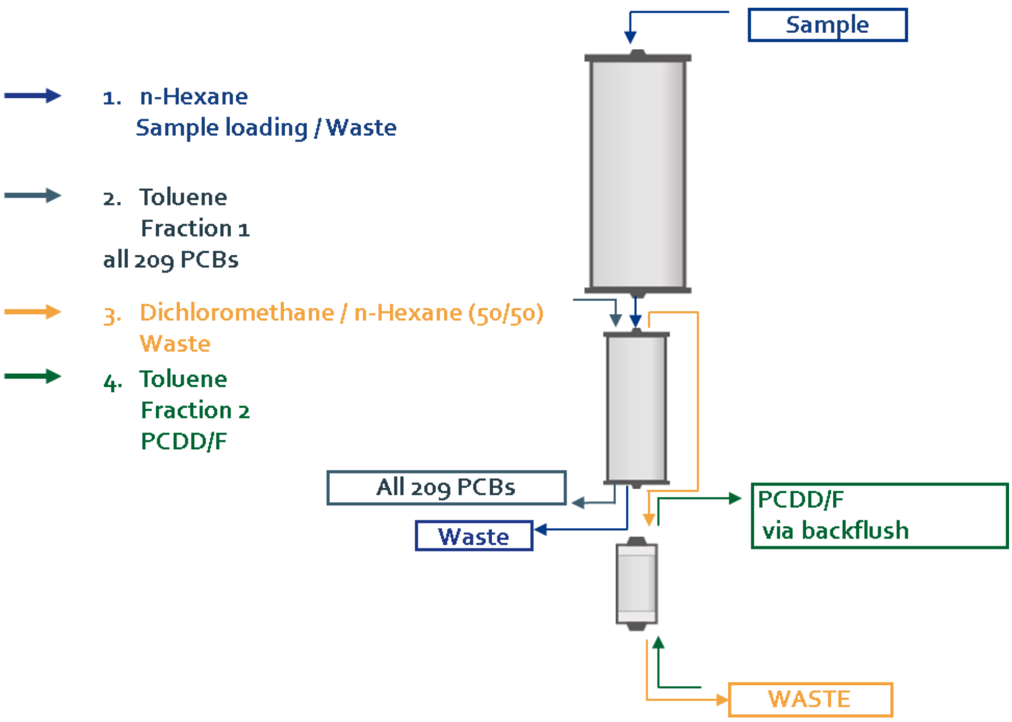
The study aimed to develop an automated clean-up method with our DEXTech system for all 209 PCBs in accordance with QC criteria of EPA method 1668C (Table 6), while also addressing the simultaneous clean-up and separation of PCDD/Fs. Unlike traditional EPA methods EPA 1613B or 1668C that focus solely on either PCBs or PCDD/Fs, this automated approach offers a comprehensive solution for both contaminants, potentially enhancing production efficiency and reducing costs in laboratory settings. Initial recovery and precision assessments were conducted on a variety of sample types, including blank samples, soil samples A and B, and fish oil samples. Thereby using LCTech systems like the X-TRACTION® system, the D-EVA system and our automated clean-up systems DEXTech Heat or DEXTech Pure for the whole workflow.

The results demonstrated compliance with established quality control (QC) criteria for labeled compounds, confirming the reliability and effectiveness of the automated cleanup method. Figures 11 and 12 provide graphical representations of mean recovery rates for $^{13}C_{12}$ -labeled PCDD/Fs and PCBs, respectively, with error bars indicating relative standard deviation (RSD) values. These figures illustrate the consistency and accuracy of the method across different sample matrices. Furthermore, fortification experiments revealed satisfactory recovery rates for native PCBs and PCDD/Fs in blank, soil, and fish oil samples, as outlined in Figure 13 and Figure 14. Overall, the study highlights the feasibility and efficacy of the automated clean-up method for comprehensive analysis of PCBs and PCDD/Fs in environmental samples.

In conclusion, the developed automated clean-up method with the LCTech DEXTech Heat or Pure system presents a significant advancement in environmental analysis, offering simultaneous clean-up and separation of PCBs and PCDD/Fs. By streamlining the process and ensuring compliance with stringent QC criteria, the method holds promise for enhancing productivity and reducing costs in environmental laboratories. The successful results of the method across various sample matrices underscores its versatility and reliability. Overall, this study contributes to our ongoing efforts to improve analytical techniques for the detection and quantification of persistent organic pollutants, thereby supporting environmental monitoring and regulatory compliance initiative.

Supplementary Materials

PCB 209+Dioxin Flow scheme and Method parameter



Method Name	PCB 209 + Dioxin		N°	10
back	Conditioning			
	ml/min	min		n-Hexane
Conditioning 1	7.0	8.0		n-Hexane
Conditioning 2	1.0	0.0		n-Hexane
Conditioning 3	1.0	0.0		Toluene
Conditioning 4	1.0	0.0		DCM/n-Hexane 1:1

Fractioning:

- **Fraction 1: all 209 PCBs**
- **Fraction 2 PCDD/F**

Process time : 52 min
Without PCDD/F: 33 min

Method Name	PCB 209 + Dioxin		N°	10
back	Fraction			
	ml/min	min		n-Hexane
Pre-run C1	7.0	3.0		n-Hexane
Fraction 1-C1	7.0	0.0		n-Hexane
Pre-run F1	7.0	10.0		n-Hexane
Fraction 1	3.0	8.0		Toluene
Pre-run F2	3.0	8.0		DCM/n-Hexane 1:1
Fraction 2	1.0	11.0		Toluene
Nitrogen		1.0		

Solvent consumption:

- **N-Hexane:** **147 mL**
- **N-Hexane/DCM (50:50%):** **24 mL**
- **Toluene:** **35 mL**

Total : 171 mL

HRMS-GC Parameters and Methods:

GC and Injector conditions - PCB

Trace 1310 GC SSL Parameters	
Operating Mode:	Splitless
Injection Volume [µL]:	5
Inlet temperature [°C]:	270
Carrier Gas, Flow [mL/min]	He, 1.0
Splitless Time [min]	1
Split Flow [mL/min]	15
Septum Purge [mL/min]	5 (constant)

Trace 1310 GC Oven Parameters	Rate [°C/min]	Temperature [°C]	Hold Time [min]
initial		120	2
1	10	200	0
2	1.5	255	0
3	10	320	5

Autosampler Settings		
Injection mode	Basic	
Rapid mode	Disable	
Injection depth	Standard	
Syringe volume [µL]	10	
Needle lengthe [mm]	57	
Sampling air and filling mode	Custom	1µL air, 2µL filling volume
Sample type	Viscous	

Table 2: GC – Program 209 PCB

GC and Injector conditions - PCDD/F

Trace 1310 GC PTV Parameters

Operating Mode:	Large Volume
Injection Volume [µL]:	5
Initial Inlet temperature [°C]:	75
Carrier Gas, Flow [mL/min]	He, 1.2
Splitless Time [min]	1
Split Flow [mL/min]	100
Septum Purge [mL/min]	5 (constant)

PTV Ramp Settings

	Pressure [Psi]	Rate [°C/s]	Temperature [°C]	Time [min]	Flow [mL/min]
Injection:	---	---	---	0.2	100
Transfer:	---	5	300	1	---
Cleaning:	---	14.5	330	5	200

Trace 1310 GC Oven Parameters

	Rate [°C/min]	Temperature [°C]	Hold Time [min]
initial		120	3
1	40	215	0.5
2	2.6	260	0
3	40	300	9
4	20	320	13

Autosampler Settings

Injection mode	Basic	
Rapid mode	Disable	
Injection depth	Standard	
Syringe volume [µL]	10	
Needle lengthe [mm]	57	
Sampling air and filling mode	Custom	1µL air, 2µL filling volume
Sample type	Viscous	

Table 3: GC – Program PCDD/F

MID 209PCB

Fn-1	
1-pcb	188,0393
1-pcb	190,0366
Lock	192,9883
1L-PCB	200,0795
1L-PCB	202,0766
2-pcb	221,9998
2-pcb	223,9968
Cali	230,9851
2L-PCB	234,0406
2L-PCB	236,0376

Fn-2	
Lock	218,9856
2-pcb	221,9980
2-pcb	223,9968
2L-PCB	234,0406
2L-PCB	236,0376
3-pcb	255,9613
3-pcb	257,9585
3L-PCB	268,0016
3L-PCB	269,9987
4-pcb	289,9224
4-pcb	291,9195
4L-PCB	301,9626
4L-PCB	303,9597
Cali	304,9824

Fn-3	
Lock	242,9856
3-pcb	255,9613
3-pcb	257,9585
3L-PCB	268,0016
3L-PCB	269,9987
4-pcb	289,9224
4-pcb	291,9195
4L-PCB	301,9626
4L-PCB	303,9597
5-pcb	325,8805
5-pcb	327,8776
5L-PCB	337,9207
5L-PCB	339,9178
6-pcb	359,8415
6-pcb	361,8386
Cali	366,9792
6L-PCB	371,8817
6L-PCB	373,8788

Fn-4	
4-pcb	289,9224
4-pcb	291,9195
Lock	292,9824
4L-PCB	301,9626
4L-PCB	303,9597
5-pcb	325,8805
5-pcb	327,8776
5L-PCB	337,9207
5L-PCB	339,9178

Fn-4	
6-pcb	359,8415
6-pcb	361,8386
6L-PCB	371,8817
6L-PCB	373,8788
7-pcb	393,8025
7-pcb	395,7996
7L-PCB	405,8428
7L-PCB	407,8398
Cali	416,9761

Fn-5	
Lock	316,9824
5-pcb	325,8805
5-pcb	327,8776
5L-PCB	337,9207
5L-PCB	339,9178
6-pcb	359,8415
6-pcb	361,8386
6L-PCB	371,8817
6L-PCB	373,8788
7-pcb	393,8025
7-pcb	395,7996
7L-PCB	405,8428
7L-PCB	407,8398
8-pcb	427,7636
8-pcb	429,7606
8L-PCB	439,8032
8L-PCB	441,8003
Cali	442,9729

Fn-6	
Lock	354,9792
6-pcb	359,8415
6-pcb	361,8386
6L-PCB	371,8817
6L-PCB	373,8788
7-pcb	393,8025
7-pcb	395,7996
7L-PCB	405,8428
7L-PCB	407,8398
8-pcb	427,7636
8-pcb	429,7606
8L-PCB	439,8032
8L-PCB	441,8003
Cali	455,9762

Fn-7	
7-pcb	393,8025
7-pcb	395,7996
Lock	404,9761
7L-PCB	405,8428
7L-PCB	407,8398
8-pcb	427,7636
8-pcb	429,7606
8L-PCB	439,8032
8L-PCB	441,8003
9-pcb	461,7246
9-pcb	463,7217
9L-PCB	473,7648
9L-PCB	475,7619
10-pcb	497,6826
10-pcb	499,6797
Cali	504,9697
10L-PCB	509,7229
10L-PCB	511,7199

Table 4: MID – all 209 PCBs

MID PCDD/F

Fn-1	
4-pcb	289,9224
4-pcb	291,9195
Lock	292,9824
4L-PCB	301,9626
tcdf	303,9016
4L-PCB	303,9597
tcdf	305,8987
TCDF	315,9419
TCDF	317,9389
tcdd	319,8965
tcdd	321,8936
13C6-TCDD	325,9161
13C6-TCDD	327,9137
TCDD	331,9368
TCDD	333,9339
Cali	366,9792

Fn-2	
Lock	292,9824
tcdf	303,9016
tcdf	305,8987
TCDF	315,9419
TCDF	317,9389
tcdd	319,8965
tcdd	321,8936
5-pcb	325,8805
13C6-TCDD	325,9161
5-pcb	327,8776
13C6-TCDD	327,9137
TCDD	331,9368
TCDD	333,9339
5L-PCB	337,9207
5L-PCB	339,9178
Cali	366,9792

Fn-3	
Lock	316,9824
5-pcb	325,8805
5-pcb	327,8776
5L-PCB	337,9207
5L-PCB	339,8597
pn-cdf	339,9178
pn-cdf	341,8568
Pn-CDF	351,9000
Pn-CDF	353,8970
pn-cdd	355,8546
pn-cdd	357,8517
6-pcb	359,8415
6-pcb	361,8386
Cali	366,9792
Pn-CDD	367,8949
Pn-CDD	369,8919
6L-PCB	371,8817
6L-PCB	373,8788

Fn-4	
6-pcb	359,8415
6-pcb	361,8386
Lock	366,9792
6L-PCB	371,8817
hx-cdf	373,8207
6L-PCB	373,8788
hx-cdf	375,8178
Hx-CDF	385,8610
Hx-CDF	387,8580
hx-cdd	389,8157
hx-cdd	391,8127
Hx-CDD	401,8559
Hx-CDD	403,8530
Cali	404,9761

Fn-5	
6-pcb	359,8415
6-pcb	361,8386
Lock	366,9792
6L-PCB	371,8817
6L-PCB	373,8788
hp-cdf	407,7818
hp-cdf	409,7788
Hp-CDF	419,8220
Hp-CDF	421,8191
hp-cdd	423,7767
hp-cdd	425,7737
Hp-CDD	435,8169
Hp-CDD	437,8140
Cali	454,9729

Fn-6	
Lock	404,9761
ocdf	441,7428
ocdf	443,7398
OCDF	453,7830
OCDF	455,7801
ocdd	457,7377
ocdd	459,7348
OCDD	469,7780
OCDD	471,7750
Cali	492,9697

Table 5: MID - PCDD/F

PCDD/F Standard Solutions and Concentrations

	VER	VF:	20	VF:	20	
	EDF-CS5	EDF-5525-100x	EDF-5525-DF-Q	EDF-5526-100x	EDF-5526-DF-I	Stocksolution
	[ng/mL]	[ng/mL]	[ng/mL]	[ng/mL]	[ng/mL]	[ng/mL]
ISS 1,2,3,4-TCDD	10	---	---	50	2,5	---
ISS 1,2,3,4,6,8,9-HpCDF	10	---	---	50	2,5	---
ISS #80L	30	---	---	150	7,5	---
2,3,7,8-TCDF	10	50	2,5	---	---	---
2,3,7,8-TCDD	10	50	2,5	---	---	---
1,2,3,7,8-PnCDF	10	50	2,5	---	---	---
2,3,4,7,8-PnCDF	10	50	2,5	---	---	---
1,2,3,7,8-PnCDD	10	50	2,5	---	---	---
1,2,3,4,7,8-HxCDF	10	50	2,5	---	---	---
1,2,3,6,7,8-HxCDF	10	50	2,5	---	---	---
1,2,3,7,8,9-HxCDF	10	50	2,5	---	---	---
2,3,4,6,7,8-HxCDF	10	50	2,5	---	---	---
1,2,3,4,7,8-HxCDD	10	50	2,5	---	---	---
1,2,3,6,7,8-HxCDD	30	150	7,5	---	---	---
1,2,3,7,8,9-HxCDD	10	50	2,5	---	---	---
1,2,3,4,6,7,8-HpCDF	30	150	7,5	---	---	---
1,2,3,4,7,8,9-HpCDF	10	50	2,5	---	---	---
1,2,3,4,6,7,8-HpCDD	30	150	7,5	---	---	---
OCDF	10	50	2,5	---	---	---
OCDD	100	500	25	---	---	---
#77L	30	150	7,5	---	---	---
#81L	30	150	7,5	---	---	---
#126L	30	150	7,5	---	---	---
#169L	30	150	7,5	---	---	---
2,3,7,8-tcdf	1	---	---	---	---	17,9
2,3,7,8-tcdd	1	---	---	---	---	19,4
1,2,3,7,8-pncdf	1	---	---	---	---	81
2,3,4,7,8-pncdf	1	---	---	---	---	87,6
1,2,3,7,8-pncdd	1	---	---	---	---	73,8
1,2,3,4,7,8-hxcdf	1	---	---	---	---	89,8
1,2,3,6,7,8-hxcdf	1	---	---	---	---	78,6
1,2,3,7,8,9-hxcdf	1	---	---	---	---	83,9
2,3,4,6,7,8-hxcdf	1	---	---	---	---	80,3
1,2,3,4,7,8-hxcdd	1	---	---	---	---	88,5
1,2,3,6,7,8-hxcdd	10	---	---	---	---	99,1
1,2,3,7,8,9-hxcdd	1	---	---	---	---	85,4
1,2,3,4,6,7,8-hpcdf	10	---	---	---	---	76,1
1,2,3,4,7,8,9-hpcdf	1	---	---	---	---	80,6
1,2,3,4,6,7,8-hpcdd	10	---	---	---	---	78,7
ocdf	1	---	---	---	---	146
ocdd	100	---	---	---	---	173
#77	10	---	---	---	---	---
#81	10	---	---	---	---	---
#126	10	---	---	---	---	---
#169	10	---	---	---	---	---
2,3,7,8-[37Cl4]-TCDD	40	---	---	---	---	---

Table 6: Standard solutions PCDD/F

PCB Standard Solutions and Concentrations

		VF: 20		VF: 20			VF: 5	
	VER ng/mL	PCB-LCS-H ng/mL	PCB-Q ng/mL	PCB-ISS-H ng/mL	PCB-I ng/mL	PCB-SCS-H ng/mL	209AG ng/mL	209AG VF5 ng/mL
PCB-ISS-#79L	50	---	---	1000	50	---	---	---
PCB-ISS-#37L	50	---	---	1000	50	---	---	---
PCB-CU-#60L	---	---	---	---	---	1000	---	---
PCB-ISS-#111L	50	---	---	1000	50	---	---	---
PCB-ISS-#162L	50	---	---	1000	50	---	---	---
PCB-CU-#159L	---	---	---	---	---	1000	---	---
PCB-#28L	50	1000	50	---	---	---	---	---
PCB-#52L	50	1000	50	---	---	---	---	---
PCB-#77L	50	1000	50	---	---	---	---	---
PCB-#81L	50	1000	50	---	---	---	---	---
PCB-#101L	50	1000	50	---	---	---	---	---
PCB-#123L	50	1000	50	---	---	---	---	---
PCB-#118L	50	1000	50	---	---	---	---	---
PCB-#114L	50	1000	50	---	---	---	---	---
PCB-#105L	50	1000	50	---	---	---	---	---
PCB-#126L	50	1000	50	---	---	---	---	---
PCB-#153L	50	1000	50	---	---	---	---	---
PCB-#138L	50	1000	50	---	---	---	---	---
PCB-#167L	50	1000	50	---	---	---	---	---
PCB-#156L	50	1000	50	---	---	---	---	---
PCB-#157L	50	1000	50	---	---	---	---	---
PCB-#169L	50	1000	50	---	---	---	---	---
PCB-#180L	50	1000	50	---	---	---	---	---
PCB-#189L	50	1000	50	---	---	---	---	---
pcb-#1 to #209	50	---	---	---	---	---	5000	1000

Table 7: Standard solutions PCB

Limit of Detection / Retentiontime

PCB	pg absolute	[min]
#1	0,02	11,26
#2	0,03	12,49
#3	0,03	12,63
#10	0,63	12,99
#4	0,82	13,06
#9	0,16	13,88
#7	0,14	13,96
#6	0,14	14,44
#8/5	0,07	14,67
#14	0,16	15,25
#11	0,26	16,2
#13/12	0,13	16,49
#15	0,25	16,79
#19	0,05	15,16
#30	0,04	15,47
#18	0,05	16,13
#17	0,06	16,29
#24	0,04	16,55
#27	0,04	16,69
#32	0,04	17,04
#16	0,06	17,16
#23	0,06	17,54
#34	0,06	17,65
#29	0,06	17,78
#26	0,06	18,08
#25	0,06	18,27
#31	0,06	18,46
#28	0,05	18,68
#21	0,04	19,14
#20/33	0,02	19,3
#22	0,04	19,68
#36	0,04	20,09
#39	0,04	20,55
#38	0,05	21,29
#35	0,04	22,05
#37	0,04	22,59
#54	0,04	17,4
#50	0,07	18,1
#53	0,07	18,68
#51	0,06	18,99
#45	0,05	19,37
#46	0,05	19,95
#52/69	0,02	20,19
#73	0,03	20,3
#43/49	0,02	20,45
#65/75/47/48	0,01	20,76
#62	0,03	20,93
#44	0,05	21,59
#59	0,03	21,7
#42	0,05	21,88
#64/72/71/41	0,01	22,34
#68	0,03	22,66
#40	0,1	23,11
#57	0,04	23,26
#67	0,04	23,66
#63/58	0,02	23,9
#61	0,05	24,19
#74	0,04	24,3
#70	0,04	24,63
#76	0,03	24,78
#80/66	0,02	24,98
#55	0,02	25,63
#60/56	0,01	26,35
#79	0,03	27,56
#78	0,03	28,49
#81	0,03	29,29
#77	0,03	30,39

PCB	pg absolute	[min]
#104	0,04	20,69
#96	0,04	22,05
#103	0,05	22,46
#100	0,06	22,94
#94	0,07	23,38
#102/93/98/95	0,02	24,03
#88	0,08	24,37
#91	0,08	24,54
#121	0,04	24,65
#84/92	0,02	25,69
#89	0,04	25,97
#90	0,04	26,18
#101	0,04	26,22
#113	0,03	26,47
#99	0,04	26,69
#112/119	0,02	27,3
#83	0,06	27,54
#108	0,02	27,62
#86/97/117	0,01	28,06
#116/125	0,02	28,2
#87/115	0,02	28,42
#111	0,03	28,67
#85	0,04	28,88
#110/120	0,02	29,26
#82	0,05	30,37
#124	0,04	31,34
#107/109	0,02	31,65
#123	0,04	31,9
#106	0,03	32,11
#118	0,03	32,27
#114	0,04	32,98
#122	0,04	33,49
#105	0,03	34,8
#127	0,03	34,97
#126	0,03	38,41
#155	0,06	24,62
#150	0,05	26,18
#152	0,05	26,74
#145	0,05	27,34
#136	0,05	27,88
#148	0,06	28,01
#154	0,06	28,77
#151	0,07	29,57
#135	0,07	29,93
#144	0,06	30,11
#147	0,07	30,2
#149/139	0,04	30,72
#140	0,07	31,12
#143	0,07	31,43
#134	0,08	31,62
#142	0,07	32,09
#131	0,08	32,16
#133	0,07	32,33
#165	0,06	32,86
#146	0,05	33,06
#132	0,08	33,18
#161	0,05	33,33
#153	0,05	33,81
#168	0,05	33,92
#141	0,05	34,78
#137	0,05	35,39
#130	0,06	35,65
#164/163	0,02	36,19
#138	0,04	36,41
#160	0,04	36,55
#158	0,04	36,73
#129	0,06	37,04

PCB	[pg absolute]	[min]
#166	0,04	37,56
#159	0,04	38,79
#128	0,07	39,11
#162	0,04	39,32
#167	0,04	40,08
#156	0,04	42,4
#157	0,03	42,97
#169	0,03	46,88
#188	0,05	31,36
#184	0,05	32,15
#179	0,05	33,28
#176	0,06	34,06
#186	0,04	34,85
#178	0,06	36,13
#175	0,06	36,8
#182/187	0,03	37,05
#183	0,06	37,76
#185	0,07	38,71
#174	0,06	39,26
#181	0,07	39,5
#177	0,07	39,83
#171	0,07	40,53
#173	0,08	41,08
#172	0,04	42,55
#192	0,03	42,96
#180	0,04	43,45
#193	0,03	43,58
#191	0,03	44,21
#170	0,04	46,34
#190	0,03	46,78
#189	0,02	49,69
#202	0,05	38,93
#201	0,05	39,84
#204	0,04	40,09
#197	0,04	40,78
#200	0,03	42,21
#198	0,05	45,93
#199	0,04	46,03
#196	0,05	46,86
#203	0,04	46,98
#195	0,02	49,23
#194	0,01	51,16
#205	0,01	51,58
#208	0,02	47,94
#207	0,02	48,7
#206	0,03	52,34
#209	0,01	52,84

PCDD/F	[pg absolute]	[min]
2,3,7,8-TCDF	0,02	25,64
2,3,7,8-TCDD	0,01	26,01
1,2,3,7,8-PeCDF	0,06	28,28
2,3,4,7,8-PeCDF	0,05	29,2
1,2,3,7,8-PeCDD	0,09	29,36
1,2,3,4,7,8-HxCDF	0,06	32,17
1,2,3,6,7,8-HxCDF	0,06	32,31
2,3,4,6,7,8-HxCDF	0,07	33,19
1,2,3,7,8,9-HxCDF	0,05	34,19
1,2,3,4,7,8-HxCDD	0,12	33,28
1,2,3,6,7,8-HxCDD	0,13	33,4
1,2,3,7,8,9-HxCDD	0,1	33,79
1,2,3,4,6,7,8-HpCDF	0,03	35,86
1,2,3,4,7,8,9-HpCDF	0,05	38,1
1,2,3,4,6,7,8-HpCDD	0,07	37,32
1,2,3,4,6,7,8,9-OCDF	0,14	42,41
1,2,3,4,6,7,8,9-OCDD	0,17	42,15

Table 8: Limit of Detection and Retention times

PCDD/F $^{13}C_{12}$ internal standard and native recoveries

PCDD/F congenere	Solvent Blank n = 4		Fish oil n = 9 ^{13}C/ n = 4 nativ		Soil A n = 9 ^{13}C/ n = 4 nativ		Soil B n = 11 ^{13}C/ n = 11 nativ				
	mean recovery %	RSD %	mean recovery %	RSD %	mean recovery %	RSD %	mean recovery %	RSD %	X (%)	RSD (%)	compliant/non-compliant
$13C_{12}$											
13C-2,3,7,8-TCDF	80	8,5	69	7,2	79	7,4	83	6,1	31-113	35,0	compliant
13C-1,2,3,7,8-PeCDF	84	11,6	64	16,2	79	9,8	78	5,7	27-156	34,0	compliant
13C-2,3,4,7,8-PeCDF	91	9,7	64	14,4	83	7,3	84	8,2	16-279	38,0	compliant
13C-1,2,3,4,7,8-HxCDF	85	10,1	94	9,3	97	17,7	104	12,6	27-152	43,0	compliant
13C-1,2,3,6,7,8-HxCDF	90	13,7	103	10,8	105	15,3	108	11,4	30-122	35,0	compliant
13C-2,3,4,6,7,8-HxCDF	89	11,1	91	11,6	101	14,8	105	9,2	29-136	37,0	compliant
13C-1,2,3,7,8,9-HxCDF	86	9,5	84	8,2	94	9,6	94	10,1	24-157	40,0	compliant
13C-1,2,3,4,6,7,8-HpCDF	90	8,4	92	7,3	105	8,6	99	6,4	32-110	41,0	compliant
13C-1,2,3,4,7,8,9-HpCDF	80	9,1	69	16,9	96	4,1	77	6,6	28-141	40,0	compliant
13C-1,2,3,4,6,7,8,9-OCDF	82	13,4	71	16,6	99	11,3	73	17,9	n.d	n.d	compliant
13C-2,3,7,8-TCDD	81	8,6	66	13,9	81	5,5	80	5,5	28-134	37,0	compliant
13C-1,2,3,7,8-PeCDD	83	10,3	62	13,7	82	12,6	79	8,3	27-184	39,0	compliant
13C-1,2,3,4,7,8-HxCDD	97	8,5	104	9,7	108	11,8	110	7,2	29-147	41,0	compliant
13C-1,2,3,6,7,8-HxCDD	79	7,4	92	8,2	93	12,7	96	13,1	34-122	38,0	compliant
13C-1,2,3,7,8,9-HxCDD	86	7,7	94	10,6	102	11,5	99	9,4	n.d	n.d	compliant
13C-1,2,3,4,6,7,8-HpCDD	85	11,5	78	10,7	102	5,1	85	4,7	34-129	35,0	compliant
13C-1,2,3,4,6,7,8,9-OCDD	77	11,1	64	18,6	93	8,6	68	13,5	20-138	48,0	compliant
nativ											
2,3,7,8-TCDF	95	2,7	101	2,9	100	1,1	88	5,4	87-137	20,0	compliant
1,2,3,7,8-PeCDF	95	1,5	111	4,8	107	3,3	90	4,8	86-124	15,0	compliant

2,3,4,7,8-PeCDF	99	3,4	107	2,7	103	0,7	91	9,1	72-150	17,2	compliant
1,2,3,4,7,8-HxCDF	99	1,9	105	3,9	117	4,0	95	7,7	82-118	17,4	compliant
1,2,3,6,7,8-HxCDF	95	2,4	103	1,6	110	2,8	94	6,5	92-120	13,4	compliant
2,3,4,6,7,8-HxCDF	100	2,5	99	2,8	107	2,1	90	9,1	84-122	12,8	compliant
1,2,3,7,8,9-HxCDF	92	4,8	100	2,1	114	1,9	98	10,0	74-148	14,8	compliant
1,2,3,4,6,7,8-HpCDF	105	0,9	110	4,2	116	2,1	98	4,4	90-130	12,6	compliant
1,2,3,4,7,8,9-HpCDF	94	3,4	117	5,2	107	2,7	93	8,4	86-126		compliant
1,2,3,4,6,7,8,9-OCDF	96	5,8	104	2,8	107	1,2	85	10,6	74-127	27,0	compliant
2,3,7,8-TCDD	101	2,9	110	2,3	102	1,6	89	2,9	83-129	28,0	compliant
1,2,3,7,8-PeCDD	106	3,2	110	6,4	109	8,2	90	6,8	76-132	15,0	compliant
1,2,3,4,7,8-HxCDD	95	1,5	102	4,9	107	2,3	82	7,3	78-152	18,8	compliant
1,2,3,6,7,8-HxCDD	106	4,7	108	1,8	114	1,5	95	4,3	84-124	15,4	compliant
1,2,3,7,8,9-HxCDD	93	2,5	107	5,7	100	1,9	85	9,6	74-142	22,2	compliant
1,2,3,4,6,7,8-HpCDD	104	2,0	114	3,5	116	1,9	98	2,9	76-130	15,4	compliant
1,2,3,4,6,7,8,9-OCDD	98	1,4	106	2,4	109	2,1	93	6,9	89-127	19,0	compliant

Table 9 shows all ^{13}C and native PCDD/F recoveries

PCB ^{13}C Internal Standard and Native Recoveries

	^{13}C internal standard and native Recoveries								IPR Criteria EPA 1668C Table 6		
209 PCBs	Solvent Blank n = 4		Fish oil n = 9 ^{13}C/ n = 4 native		Soil A n = 9 ^{13}C/ n = 4 native		Soil B n = 11 ^{13}C/ n = 11 nativ				
	mean recovery %	RSD %	mean recovery %	RSD %	mean recovery %	RSD %	mean recovery %	RSD %	mean recovery %	RSD (%)	compliant/non-compliant
$^{13}C_{12}$											
#1L	104	2,0	83	18,5	102	19,8	65	21,1	20-135	70,0	compliant
#3L	98	1,4	73	12,8	103	18,9	62	26,9	20-135	70,0	compliant
#4L	78	13,2	56	7,9	73	10,2	40	27,9	20-135	70,0	non compliant
#8L	70	7,2	73	5,8	84	2,3	75	12,7	20-135	70,0	compliant
#15L	47	3,0	67	14,2	75	8,7	72	20,8	20-135	70,0	compliant
#19L	75	3,7	69	14,9	83	7,5	83	13,2	20-135	70,0	compliant
#28L	71	7,3	84	6,8	102	7,2	78	17,3	20-135	70,0	compliant
#37	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	20-135	70,0	
#54L	80	8,0	77	9,3	98	10,3	73	21,1	20-135	70,0	compliant
#52L	84	9,9	96	11,0	110	8,4	89	14,0	45-135	50,0	compliant
#70L	84	6,1	94	7,9	116	11,8	95	9,3	45-135	50,0	compliant
#60L	101	1,4	100	0,0	100	0,0	100	0,0	45-135	50,0	compliant
#81L	76	2,9	81	7,3	89	4,3	93	13,5	45-135	50,0	compliant
#77L	78	4,0	78	8,3	88	7,7	99	6,7	45-135	50,0	compliant
#104L	81	8,8	92	15,9	113	11,5	84	13,1	45-135	50,0	compliant
#95L	80	5,4	82	11,4	111	13,4	78	11,1	45-135	50,0	compliant
#101L	77	3,6	81	8,3	93	8,1	78	20,1	45-135	50,0	compliant
#123L	78	2,6	72	6,3	81	9,8	83	15,0	45-135	50,0	compliant

#118L	76	3,9	67	4,7	78	8,6	83	15,8	45-135	50,0	compliant
#114L	78	2,9	74	4,7	80	9,8	91	22,7	45-135	50,0	compliant
#105L	78	4,3	67	6,5	73	11,1	81	12,9	45-135	50,0	compliant
#126L	73	3,0	74	16,7	81	17,4	94	5,0	45-135	50,0	compliant
#155L	68	6,9	91	11,5	110	17,2	80	10,7	45-135	50,0	compliant
#153L	80	7,3	83	11,2	98	10,3	80	10,5	45-135	50,0	compliant
#138L	78	5,6	71	7,6	84	6,3	84	9,9	45-135	50,0	compliant
#159L	101	1,5	100	0,0	100	0,0	100	0,0	45-135	50,0	compliant
#167L	79	3,3	76	5,2	88	2,4	87	5,4	45-135	50,0	compliant
#156L	77	3,4	75	11,9	85	4,7	84	8,3	45-135	50,0	compliant
#157L	78	5,7	76	12,7	91	5,1	88	9,0	45-135	50,0	compliant
#169L	81	4,2	103	12,3	109	5,9	126	8,4	45-135	50,0	compliant
#180L	79	4,1	84	9,4	97	6,7	86	9,0	45-135	50,0	compliant
#170L	78	3,2	87	9,2	104	18,1	96	15,1	45-135	50,0	compliant
#188L	71	7,0	84	16,2	103	16,2	78	14,5	45-135	50,0	compliant
#189L	74	4,5	83	12,6	102	15,1	85	10,3	45-135	50,0	compliant
#202L	73	6,5	66	16,9	71	15,9	71	11,6	45-135	50,0	compliant
#205L	76	4,5	74	6,3	87	1,8	84	6,1	45-135	50,0	compliant
#206L	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	45-135	50,0	compliant
#208L	63	5,7	75	4,2	73	9,9	76	12,0	45-135	50,0	compliant
#209L	46	10,9	72	8,0	67	12,1	79	11,5	45-135	50,0	compliant
nativ											
#1	73	8,5	105	1,8	113	1,8	102	6,1	70-130	25	compliant
#2	80	11,0	121	1,7	106	2,5	110	12,8	70-130	25	compliant
#3	74	11,1	108	2,6	100	2,4	101	4,9	70-130	25	compliant
#10	105	10,2	122	6,6	125	4,5	108	24,7	70-130	25	compliant
#4	86	8,0	117	3,6	112	2,2	103	5,0	70-130	25	compliant
#9	84	12,5	114	1,8	109	2,4	98	7,3	70-130	25	compliant
#7	82	12,3	126	2,6	117	2,1	98	9,3	70-130	25	compliant

#6	85	8,4	116	2,2	110	3,3	99	9,3	70-130	25	compliant
#8/5	84	5,1	103	1,9	101	2,5	100	6,7	70-130	25	compliant
#14	88	5,7	101	1,6	92	2,1	100	7,0	70-130	25	compliant
#11	88	9,4	106	3,3	97	3,9	119	16,8	70-130	25	compliant
#13/12	87	7,9	109	3,5	104	1,7	109	12,0	70-130	25	compliant
#15	87	10,9	111	3,9	101	2,6	98	6,9	70-130	25	compliant
#19	83	11,6	109	2,6	111	1,0	104	1,9	70-130	25	compliant
#30	89	6,6	113	2,6	109	0,8	100	18,5	70-130	25	compliant
#18	90	5,6	100	3,6	103	1,1	99	14,2	70-130	25	compliant
#17	91	5,5	104	2,8	105	1,2	104	14,1	70-130	25	compliant
#24	90	4,5	103	3,0	103	1,1	104	3,1	70-130	25	compliant
#27	90	4,8	101	1,9	103	1,1	106	5,1	70-130	25	compliant
#32	90	4,0	103	2,9	104	1,4	100	13,9	70-130	25	compliant
#16	88	6,5	102	3,5	106	1,1	95	18,0	70-130	25	compliant
#23	99	6,1	119	2,6	117	1,6	107	17,0	70-130	25	compliant
#34	103	6,4	112	2,8	112	1,4	107	11,2	70-130	25	compliant
#29	99	7,0	119	3,7	116	1,8	112	13,1	70-130	25	compliant
#26	98	6,7	113	2,4	112	1,6	112	11,0	70-130	25	compliant
#25	90	8,6	116	2,5	114	1,7	115	17,0	70-130	25	compliant
#31	95	4,5	114	3,4	114	1,8	113	9,4	70-130	25	compliant
#28	93	4,8	106	3,1	107	1,6	106	8,0	70-130	25	compliant
#21	96	5,2	115	2,8	116	1,3	95	20,0	70-130	25	compliant
#20/33	99	3,4	97	2,8	100	2,1	95	17,2	70-130	25	compliant
#22	94	3,9	98	4,1	93	2,8	95	18,9	70-130	25	compliant
#36	97	3,4	107	4,2	102	3,5	105	20,3	70-130	25	compliant
#39	94	4,2	102	3,7	90	3,0	105	23,5	70-130	25	compliant
#38	96	5,1	106	3,7	101	3,6	105	23,4	70-130	25	compliant
#35	100	2,4	99	4,8	96	3,5	99	6,2	70-130	25	compliant
#37	99	4,1	93	4,5	88	3,8	108	23,4	70-130	25	compliant

#54	92	6,5	113	1,8	115	0,4	110	2,8	70-130	25	compliant
#50	98	5,4	125	2,8	130	2,9	113	2,4	70-130	25	compliant
#53	94	6,4	118	1,6	123	2,9	108	10,6	70-130	25	compliant
#51	95	5,2	105	2,7	121	2,9	95	10,2	70-130	25	compliant
#45	96	6,4	118	1,9	125	3,1	101	14,1	70-130	25	compliant
#46	96	6,1	120	1,6	122	2,8	106	2,8	70-130	25	compliant
#52/69	101	3,0	107	2,4	107	1,8	109	4,0	70-130	25	compliant
#73	101	4,4	94	3,4	118	2,8	106	6,3	70-130	25	compliant
#43/49	102	3,6	119	1,6	116	2,8	114	6,2	70-130	25	compliant
#65/75/47/48	102	3,3	105	2,1	102	1,4	110	3,8	70-130	25	compliant
#62	100	3,4	115	2,4	119	1,4	114	10,5	70-130	25	compliant
#44	103	2,0	126	2,3	124	2,0	112	18,6	70-130	25	compliant
#59	103	3,2	120	2,3	119	1,6	115	18,4	70-130	25	compliant
#42	104	3,6	128	1,7	128	2,1	109	15,5	70-130	25	compliant
#64/72/71/41	102	1,9	118	1,8	114	1,6	115	21,6	70-130	25	compliant
#68	101	1,8	117	2,5	115	1,0	109	2,8	70-130	25	compliant
#40	104	5,1	118	0,9	123	1,4	115	11,0	70-130	25	compliant
#57	104	5,2	127	1,5	123	1,1	119	13,5	70-130	25	compliant
#67	101	4,0	110	1,9	120	1,9	107	9,0	70-130	25	compliant
#63/58	100	6,3	123	2,7	124	1,0	104	12,0	70-130	25	compliant
#61	98	11,6	114	2,4	123	2,2	105	10,1	70-130	25	compliant
#74	101	5,3	106	3,4	116	2,1	117	5,3	70-130	25	compliant
#70	98	6,6	114	2,4	116	2,0	110	4,0	70-130	25	compliant
#76	101	4,3	121	2,4	121	2,4	100	3,0	70-130	25	compliant
#80/66	100	5,8	87	2,6	87	1,5	107	9,8	70-130	25	compliant
#55	98	4,0	103	8,6	84	4,1	85	20,0	70-130	25	compliant
#60/56	92	8,8	102	9,8	80	1,8	84	24,3	70-130	25	compliant
#79	89	2,2	114	8,4	128	2,4	90	2,9	70-130	25	compliant
#78	86	4,1	104	3,0	97	1,9	93	9,7	70-130	25	compliant

#81	89	2,4	104	4,1	93	1,2	88	8,8	70-130	25	compliant
#77	85	3,7	96	2,8	90	2,0	88	10,3	70-130	25	compliant
#104	98	2,5	112	3,9	112	1,3	108	4,5	70-130	25	compliant
#96	97	4,0	109	4,5	113	2,4	108	4,0	70-130	25	compliant
#103	101	4,4	109	5,0	109	2,1	117	8,1	70-130	25	compliant
#100	101	2,7	113	1,0	120	1,2	119	6,2	70-130	25	compliant
#94	102	2,0	108	1,3	118	1,4	119	6,7	70-130	25	compliant
#102/93/98/95	103	2,2	117	0,7	106	1,4	115	5,5	70-130	25	compliant
#88	102	2,8	128	1,2	116	2,0	122	9,9	70-130	25	compliant
#91	111	6,1	118	2,3	118	0,9	124	17,8	70-130	25	compliant
#121	98	2,5	118	1,6	115	2,3	126	12,1	70-130	25	compliant
#84/92	109	2,9	97	6,2	109	21,6	97	19,4	70-130	25	compliant
#89	104	5,7	122	2,7	111	1,3	112	9,9	70-130	25	compliant
#90	102	8,7	120	8,7	118	5,8	97	6,2	70-130	25	compliant
#101	106	8,3	101	11,6	114	2,4	122	12,7	70-130	25	compliant
#113	105	4,6	115	2,0	125	2,7	114	4,9	70-130	25	compliant
#99	104	3,1	121	6,7	127	3,1	109	13,4	70-130	25	compliant
#112/119	102	2,2	109	3,0	126	3,5	111	7,4	70-130	25	compliant
#83	105	4,5	107	8,0	115	5,6	112	5,4	70-130	25	compliant
#108	103	3,4	97	3,1	108	2,4	113	4,4	70-130	25	compliant
#86/97/117	100	5,5	96	4,7	108	3,2	103	13,1	70-130	25	compliant
#116/125	100	7,2	101	4,5	113	4,1	100	4,1	70-130	25	compliant
#87/115	98	8,5	98	5,2	108	4,0	105	6,9	70-130	25	compliant
#111	100	8,7	99	3,7	103	4,9	117	15,5	70-130	25	compliant
#85	101	4,9	98	6,1	108	4,2	112	12,9	70-130	25	compliant
#110/120	101	1,8	77	7,2	96	4,0	116	16,3	70-130	25	compliant
#82	101	2,5	85	4,0	95	4,7	114	6,4	70-130	25	compliant
#124	101	3,5	125	1,6	123	1,6	112	6,2	70-130	25	compliant
#107/109	102	4,5	116	2,1	117	2,0	112	4,9	70-130	25	compliant

#123	105	12,4	121	3,0	125	3,2	95	9,3	70-130	25	compliant
#106	113	12,1	123	2,1	123	1,9	96	10,2	70-130	25	compliant
#118	118	9,3	127	7,4	117	3,7	110	24,1	70-130	25	compliant
#114	100	2,0	130	1,4	118	2,5	110	3,7	70-130	25	compliant
#122	100	2,9	115	2,3	121	2,6	105	11,5	70-130	25	compliant
#105	98	6,6	121	2,5	114	3,2	106	6,9	70-130	25	compliant
#127	99	8,2	125	5,7	125	3,6	130	8,8	70-130	25	compliant
#126	88	3,0	94	1,4	93	2,1	91	4,8	70-130	25	compliant
#155	102	1,6	112	2,2	113	1,6	113	3,0	70-130	25	compliant
#150	114	14,3	105	4,1	109	4,0	106	11,2	70-130	25	compliant
#152	111	14,8	110	4,3	112	4,9	98	9,7	70-130	25	compliant
#145	113	14,8	100	4,3	112	5,3	102	12,6	70-130	25	compliant
#136	110	10,4	101	6,6	109	5,7	98	8,6	70-130	25	compliant
#148	121	11,4	96	4,7	101	3,9	105	7,7	70-130	25	compliant
#154	112	13,2	89	5,7	95	6,4	104	11,9	70-130	25	compliant
#151	113	13,8	74	15,2	95	5,9	89	6,6	70-130	25	compliant
#135	114	12,8	92	8,7	96	4,3	99	5,7	70-130	25	compliant
#144	111	11,5	95	5,9	90	7,5	102	11,1	70-130	25	compliant
#147	115	18,2	96	8,4	97	5,9	116	10,2	70-130	25	compliant
#149/139	115	13,8	71	20,5	92	5,7	82	9,0	70-130	25	compliant
#140	103	7,4	105	0,8	118	1,6	106	10,1	70-130	25	compliant
#143	102	8,1	107	1,1	118	1,3	109	6,9	70-130	25	compliant
#134	103	7,5	102	1,8	115	2,5	108	13,0	70-130	25	compliant
#142	90	11,6	97	4,2	112	3,0	104	7,7	70-130	25	compliant
#131	117	16,8	108	3,7	118	2,2	107	6,2	70-130	25	compliant
#133	106	7,6	103	2,2	114	1,4	120	19,4	70-130	25	compliant
#165	103	3,5	105	2,2	109	2,5	108	13,1	70-130	25	compliant
#146	99	5,2	110	7,6	97	3,4	98	17,1	70-130	25	compliant
#132	109	4,9	110	5,9	128	3,1	105	24,1	70-130	25	compliant

#161	106	6,8	109	1,2	112	1,0	106	8,2	70-130	25	compliant
#153	98	5,7	102	6,1	109	2,5	119	7,1	70-130	25	compliant
#168	106	5,2	92	5,2	113	2,5	90	26,7	70-130	25	compliant
#141	99	9,0	111	2,1	112	5,6	105	7,7	70-130	25	compliant
#137	103	2,4	120	5,4	122	2,0	107	15,0	70-130	25	compliant
#130	103	3,9	118	3,7	122	1,7	105	9,9	70-130	25	compliant
#164/163	103	2,5	106	5,7	111	1,3	104	9,6	70-130	25	compliant
#138	104	4,1	88	15,6	95	1,7	114	10,7	70-130	25	compliant
#160	104	4,3	91	17,2	109	2,0	103	6,8	70-130	25	compliant
#158	106	7,0	95	4,7	119	2,1	111	7,8	70-130	25	compliant
#129	101	7,7	128	4,3	119	2,1	104	13,2	70-130	25	compliant
#166	99	3,1	117	4,4	118	3,3	105	10,0	70-130	25	compliant
#159	104	2,7	123	4,9	123	4,3	105	10,0	70-130	25	compliant
#128	122	6,9	116	6,7	118	4,9	118	17,8	70-130	25	compliant
#162	96	11,5	115	3,3	117	2,6	106	4,3	70-130	25	compliant
#167	100	3,6	110	1,6	113	0,8	109	2,9	70-130	25	compliant
#156	103	3,8	109	4,3	103	0,8	104	6,4	70-130	25	compliant
#157	103	4,1	111	1,3	107	1,4	103	4,2	70-130	25	compliant
#169	91	4,9	95	3,1	93	1,9	91	3,5	70-130	25	compliant
#188	104	2,2	113	1,7	111	1,7	112	3,5	70-130	25	compliant
#184	101	4,9	105	0,8	107	2,0	108	6,5	70-130	25	compliant
#179	115	11,4	113	0,9	117	4,0	103	12,6	70-130	25	compliant
#176	112	12,2	110	0,8	117	4,9	115	9,6	70-130	25	compliant
#186	109	10,9	115	1,3	113	7,3	114	17,8	70-130	25	compliant
#178	114	9,6	105	4,1	106	8,5	107	14,8	70-130	25	compliant
#175	114	10,1	102	4,1	104	9,3	114	15,7	70-130	25	compliant
#182/187	113	9,2	113	10,6	108	10,4	92	17,9	70-130	25	compliant
#183	113	8,9	112	8,7	107	10,5	104	14,3	70-130	25	compliant
#185	111	8,7	108	6,7	108	12,6	107	11,9	70-130	25	compliant

#174	114	9,8	112	9,2	111	10,7	84	10,9	70-130	25	compliant
#181	114	10,0	112	9,2	112	11,6	112	15,7	70-130	25	compliant
#177	116	10,1	119	10,4	117	11,7	93	14,4	70-130	25	compliant
#171	115	10,8	116	9,3	115	14,6	110	12,3	70-130	25	compliant
#173	105	2,7	101	6,9	115	5,7	108	8,1	70-130	25	compliant
#172	104	2,3	108	3,7	111	2,7	107	3,3	70-130	25	compliant
#192	103	2,6	104	2,7	110	3,0	110	3,7	70-130	25	compliant
#180	101	9,5	95	6,3	112	2,6	101	14,3	70-130	25	compliant
#193	105	4,5	106	3,6	122	1,2	97	14,3	70-130	25	compliant
#191	102	2,3	118	3,0	120	1,6	111	4,7	70-130	25	compliant
#170	102	2,9	112	2,6	112	1,1	103	11,2	70-130	25	compliant
#190	98	2,5	120	1,4	126	7,9	102	9,5	70-130	25	compliant
#189	100	1,8	106	4,3	100	1,1	106	4,9	70-130	25	compliant
#202	104	2,7	111	1,6	112	1,1	108	4,0	70-130	25	compliant
#201	108	5,3	114	3,0	116	1,8	116	9,8	70-130	25	compliant
#204	94	13,9	115	2,3	102	5,3	115	9,4	70-130	25	compliant
#197	106	7,8	120	2,2	117	2,5	119	17,7	70-130	25	compliant
#200	113	4,7	126	4,4	122	6,2	109	3,1	70-130	25	compliant
#198	110	5,8	128	7,5	123	5,8	112	5,5	70-130	25	compliant
#199	116	9,3	112	6,3	122	3,5	108	5,2	70-130	25	compliant
#196	112	9,2	126	6,5	107	8,9	117	7,7	70-130	25	compliant
#203	107	11,8	110	5,9	125	6,5	114	4,7	70-130	25	compliant
#195	108	2,9	114	2,4	110	2,6	106	11,4	70-130	25	compliant
#194	106	2,2	112	3,3	111	2,1	103	4,2	70-130	25	compliant
#205	102	2,2	111	2,2	109	0,4	107	2,8	70-130	25	compliant
#208	104	2,1	111	0,8	111	0,8	109	3,6	70-130	25	compliant
#207	99	4,1	109	1,6	107	1,5	107	4,6	70-130	25	compliant
#206	105	12,1	109	2,5	126	5,5	114	11,8	70-130	25	compliant
#209	104	2,9	114	1,0	111	1,4	109	2,3	70-130	25	compliant

Table 10 shows all the recoveries of $^{13}C_{12}$ labeled PCBs as well as the native recoveries. In addition the compliance with the QC criteria of EPA method 1668C Table 6 is shown.

The green colour highlights the $^{13}C_{12}$ -PCBs present in our $^{13}C_{12}$ -PCB standard but not included in the standard outlined in EPA method 1668C.
Conversely, the red colour indicates the absence of certain $^{13}C_{12}$ -PCBs in our $^{13}C_{12}$ standard compared to the EPA method 1668C standard.
The blue colour marks native PCBs for which concentrations and recoveries could be calculated using a matching $^{13}C_{12}$ C-labeled standard.

Instruments & Clean-up Columns

Instruments

1.	DEXTech Pure	p/n	15969
2.	DEXTech Heat	p/n	16800
3.	DEXTech 16	p/n	15430
4.	D-EVA	p/n	16900
5.	X-TRACTION®	p/n	20000

Clean-Up Columns

1.	Universal Column	p/n	19511
2.	PCB 209 PP Column	p/n	20325
3.	Smart Column	p/n	19513
4.	Alox Column Glass	p/n	15433
5.	Alox Evolution PP	p/n	20087
6.	Carbon Column	p/n	20777
7.	Carbon Column PCB 209 Method	p/n	21344

Any questions?
Do not hesitate to contact us:

LCTech GmbH
Daimlerstr. 4
84419 Obertaufkirchen, Germany
Tel. +49 8082 2717-0
info@LCTech.de
www.LCTech.de