Dioxin, PCB analysis in fishmeal
Comparison of two extraction techniques

Abstract

For the extraction of dioxins and PCBs from fish and derived products such as fish meal quite often classical soxhlet extraction is used. The disadvantages of classical soxhlet extraction are well known: time consuming and the usage of large volumes of solvent. The use of automated systems based on extraction under high pressure and high temperature or so called Pressurized Liquid Extraction (PLE) is a major improvement however cost of these systems is relatively high and still the final volume is around 50-80 ml. In this prelimary study a technique introduced in 1974 by E.L Randall (1) is compared with a method based on Pressurized Liquid Extraction (PLE). Extractions are performed with fishmeal, whereas the Randall extractions are performed with a fully automated system based on the Randall principle called the Solvent ExtractoR (SER-158), introduced by Velp Scientifica

Introduction

The detection of polychlorinated dibenzo-p-dioxins (PCDDs), dibenzofurans (PCDFs) and polychlorinated biphenyls (PCBs) in fishmeal is of great importance especially from fish caught in contaminated areas such as the Baltic sea. Extraction is often done by classical soxhlet giving good results, however the technique is time and solvent consuming. In general extraction takes 18 hours and uses around 200-400 ml solvent. A good alternative is the use of system based on pressurized liquid extraction (PLE). Although PLE is a fast and a useful technique, a frequently observed major drawback is blocking of tubing by high protein containing samples such as fishmeal. In order to achieve the requested low LOQs the method to be applied for fishmeal requires a sample intake of 10-15 gram at least. Using a PLE system the ratio of sample to solvent using a standard cell =< 1:3 results in poor recoveries. Even in the case of multiple extraction cycles -up to three- the recovery remained quite poor. This poor recovery has been regularly observed not only in case of fishmeal samples but also for grass, hay silage and alike.

To test the recently introduced SER-158 system, DSP-systems performed experiments to see if this new automated system could be used for routine analysis.
The SER-158 operates in subsequent extraction processes as follows (see figure 1).

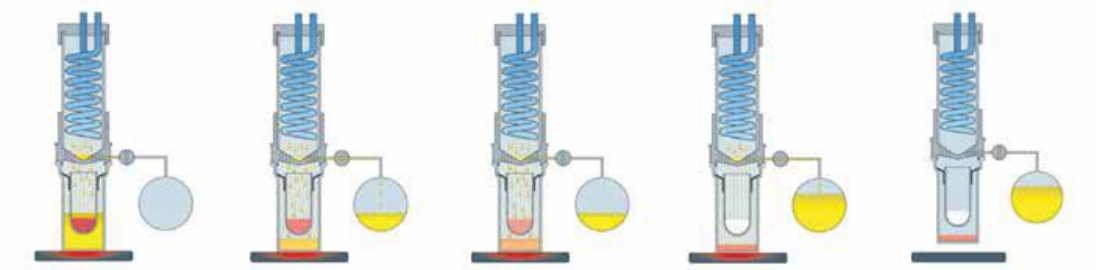

Figure 1: Five step process of SER 158 fully automated extractor

The first step is the so called “immersion” step. The sample, in a thimble, is immersed in the organic solvent which is boiled for at least one hour. During that process 80-90 % of the compounds of interest are extracted from the sample.

After typically 1 hour the system continues to the second step “removing” in which part of the extraction solvent is evaporated and transferred via a valve to the recovery tank. In case of 100 ml extraction solvent, a reproducible 50% is evaporated by means of a special developed ‘extraction cone’

The third step of the process “washing” is actually a classical hot soxhlet or twisselman extraction, assuring quantitative extraction of all compounds of interest. This step typically takes 40-60 minutes.

After the “washing” step is completed, almost all remaining solvent is removed from the glass extraction cup in the fourth step in the extraction process called “removing”. By constantly opening the valve to the solvent recovery tank all condensed solvent is removed via the cone to the solvent recovery tank. After all solvent is evaporated, heating of the samples is automatically terminated to prevent burning. The heating process is controlled individually per sample by means of sensors.

When all solvent of the last sample in the system has been evaporated, all samples are lifted above the heating plate and after 5 minutes of “cooling” the extraction process has finished.

Remark: After the sample is completely extracted and all solvent is removed the glass cup is still hot with a possible risk of evaporation of the compounds of interest or of irreversible adsorption on the glass wall. In case of fat containing samples (food and feed) the fat phase acts as a keeper minimizing this risk. In case of low or non-fat containing samples e.g. minerals, soil, sediment and alike it is advise to add 2ml dodecane to the extraction solvent . After the sample is completely extracted and all solvent is removed this 2 ml dodecane acts as keeper.

Purification of extracts

For the purification of obtained extracts the in 2015 via DSP-Systems in Europe introduced new Japanese technology (5,6) was used. The main advantages of this new techniques are:

- Low solvent consumption (100ml per sample)
- Faster (concentration of 1.5 ml extracts only)
- Easy connection of columns
- Excellent clean-up due to high performance adsorbents and heated zones
- Unique way of flow switching without valves
- No cross contamination

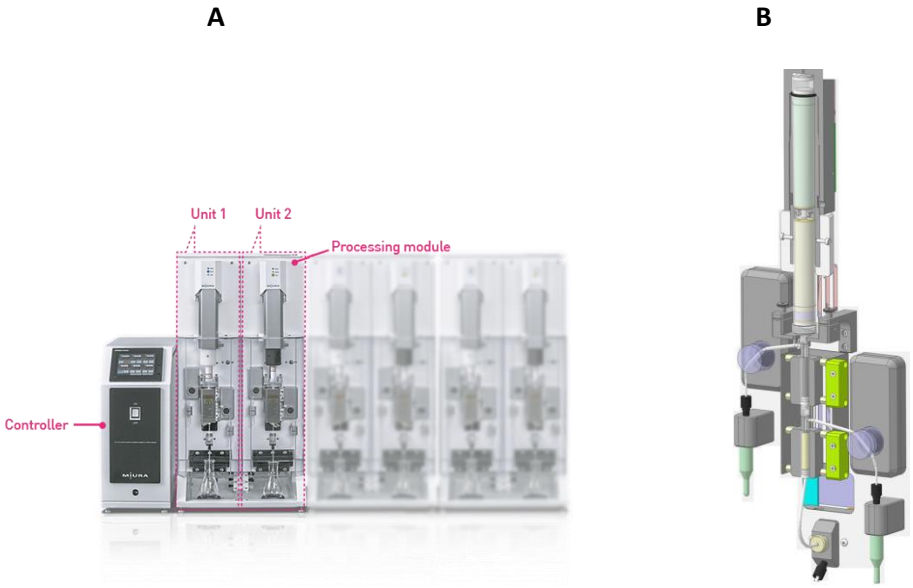

*Figure 3: **A** Set up of the GO-xHT system capable of simultaneous purifying 2, 4 or 6 samples within 90 minutes. **B** Schematic diagram of the column flow channel of the GO-xHT system.*

Materials and methods

Extraction

A reference fishmeal sample, daily used in the laboratory for QA, was in fourfold extracted using:

1) Pressurised Liquid Extraction (Speedextractor Buchi)
2) Automated Solvent Extractor based on Randall principle (SER-158 Velp Scientifica)

Ad 1. 15 gram of the sample was spiked with all relevant ^{13}C labelled congeners and mixed with hydromatrix. The mixture was transferred to the extraction cell and extracted with hexane/dichloromethane (1:1, v/v) at 100°C and 1500 PSI during 10 minutes. Followed by two additional cycles, total extraction time is 45 minutes. All extracts obtained after extraction with hexane/dichloromethane are concentrated down to 5-10 ml using a rotary evaporator. After addition of the ^{37}Cl-2,3,7,8-TCDD (clean-up standard) the final volume is made up to approximately 10 ml with hexane. Finally extracts are purified .

Ad 2. 15 gram of the sample was spiked with all relevant ^{13}C labelled congeners and mixed with hydromatrix. The mixture was transferred into a thimble and placed in the glass cups of the automated Solvent Extractor SER-158. The cups are filled via a dispenser with each 100 ml Hexane/DCM (1:1 v/v)*1 and the following five steps are automatically performed.

1. Immersion : Thimble with sample is placed in boiling solvent (60 min)
2. Removing : Solvent is evaporated until just below the thimble (10 min)*1
3. Washing : Classical hot soxhlet / twisselman extraction (60 min)
4. Recovery : Solvent is evaporated to near dryness (15 min) *1
5. Cooling : Glass cup is cooled (5 min)

To each cup 10-15 ml hexane is added as well as a clean-up standard e.g. ^{37}Cl-2,3,7,8-TCDD. Finally extracts are purified .

**1 In case of compound feed, minerals, soil or alike the extraction solvent should consist of a mixture of toluene ethanol (3:7 v/v). Times for step 2 and 4 should be increased to respectively 15 and 30 minutes. As final extracts contains toluene a solvent exchange to hexane is necessary. Solvent transfer is done by adding 5 ml methanol to the extract. The formed azeotrope (Toluene/Methanol) with a boiling point of 65°C is again evaporated . The residue is dissolved in 10 ml hexane, $^{37}Cl_4$-2,3,7,8-TCDD (clean-up standard) is added and the extracts are purified.*

Purification of extracts

Clean-up is performed using the new purification technology (GO-xHT, Miura).For each extract a set of four in-line columns is required: silica gel impregnated with silver nitrate (1); silica gel impregnated with sulfuric acid (2); activated carbon (3) and alumina (4). Column 1 and 2 are used for purification of the extracts while the other two columns are used for trapping the compounds of interest. Extracts are transferred via a funnel to the first column ($AgNO_3$ Silica). Thereafter the set of columns and tubing is assembled and placed in the GO-xHT system, figure 3B. Column set is eluted with 90 ml of hexane with a flowrate of 2.5 ml minutes. During this step the temperature of the two purification columns is maintained at 60°C. The elevated temperature weakens the adsorption with silica gel and as a result the elution speed of dioxins and PCBs is enhanced. Also the chemical reaction rates (oxidation with sulfuric acid or nitric acid) with sample matrices is accelerated. PCDD/Fs and the four NO-PCBs are trapped on the activated carbon column while the MO, NDL-PCBs and PBDEs are trapped on the alumina column.

Finally in backflush, both the alumina and the carbon column are eluted using a small amount of toluene resulting in two fractions each of 1.5 ml. During these elution steps the temperature of the carbon and alumina column is set at 90°C. At first only the alumina column is eluted and the collected fraction contains the MO-PCBs, the NDL-PCBs (and all PBDEs). After that the carbon column is eluted with toluene and this fraction contains all PCDD/Fs and NO-PCBs. To both fraction the recovery/syringe standards ^{13}C-1,2,3,4-TCDD and ^{13}C 2,3,4,6,7,8-HxCDF in 20 µl nonane is added and both fraction are concentrated to a final volume of 20 µl using an evaporator (CentriVap, Labconco).

GC-HRMS

The two obtained fractions are analysed using GC-HRMS (DFS High Resolution Magnetic Sector MS - Thermo Scientific) each MS is equipped with two GCs each with a PTV injector (Best P.T.V. injector) using a sintered glass liner (SGE pn 092155). For the determination of PCDD/Fs and the PCBs a VF-5ms 60m x 0,25mm x 0.25µm + 5m EZ-guard (Varian) GC column is used. For the determination of PBDEs a DB-5ms 15m x 0.25 mm x 0.10 µm GC columns is used. The mass spectrometer is operated in electron impact ionization mode, using selected-ion monitoring. From both fractions 4 µl is used to introduce the sample onto the GC.

Results and discussion

The new fully automated soxhlet technique "SER-158", based on Randall principles (8) can be regarded as excellent alternative for classical soxhlet extraction as well as for pressurized liquid extraction. This automated system starts with a sample intake of up to 20 gram and the required volume of organic solvent is 70 up to 100 ml. Depending on the boiling point of the organic solvent the total extraction time required is approximately 2.5 hours. As the solvent is automatically concentrated down near dryness the final extract can directly be purified using the Miura GO-xHT system. Obtained extracts can be concentrated after transfer in a GC vial with tapered end which can be placed directly in an auto-sampler.

As a proof of principle four fishmeal samples were extracted and purified using the combination of this new extraction and purification technique. In table 1 overall results of these four samples is given while in annex 1. Detailed results can be found

PCDD/Fs	Results obtained via SER158	Results obtained via PLE
Average	79.1	53.6
St. dev	9.3	13.8
CV (%)	11.7	25.7
PCBs		
Average	72.3	47.6
St. dev	9.6	22.9
CV (%)	13.3	48.1

Table 1: Overall results of recovery of 13C labeled internal standards

From the first experiments it can be concluded that the SER-158 in combination with a MIURA purification system gives good results

By using the SER-158 extraction systems, a GO-6HT purification system and a Centrivap concentrator, one technician can perform the complete sample preparation procedure of more than 100 Food and/or Feed samples in one week time.

References

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Annex 1:

Detailed results fo six samples comparison of extraction SER158 versus PLE

DIOXINS

Extraction method	**SER158**					**SER158**					**PLE**				
Sample intake g.	20.009					20.0054					20.0582				
Moisture (%)	7.0					7.0					7.0				
Sample code:	SER158-1					SER158-2					PLE 1 +2 *1				
Compound Name	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ
2378-TCDD	80	0.08	0	0.08	0.08	84	0.07	0.01	0.07	0.07	40	0.12	0	0.12	0.12
12378-PeCDD	87	0.06	0.01	0.06	0.06	92	0.06	0.01	0.06	0.06	46	0.08	0.01	0.08	0.08
123478-HxCDD	58	0.01	0.01	0.00	0.00	67	n.d.	0.01	0.00	n.d.	37	0.03	0.01	0.00	0.00
123678-HxCDD	80	0.19	0.01	0.02	0.02	85	0.20	0.01	0.02	0.02	44	0.27	0.01	0.03	0.03
123789-HxCDD		0.04	0.01	0.00	0.00		0.03	0.01	0.00	0.00		0.08	0.01	0.01	0.01
1234678-HpCDD	79	0.16	0.01	0.00	0.00	84	0.17	0.01	0.00	0.00	42	0.25	0.01	0.00	0.00
OCDD	68	0.10	0.01	0.00	0.00	72	0.11	0.01	0.00	0.00	30	0.23	0.02	0.00	0.00
2378-TCDF	75	2.62	0.02	0.26	0.26	78	2.63	0.01	0.26	0.26	39	3.21	0.02	0.32	0.32
12378-PeCDF	78	0.49	0.01	0.01	0.01	85	0.48	0.01	0.01	0.01	41	0.65	0.01	0.02	0.02
23478-PeCDF	80	0.38	0.01	0.11	0.11	88	0.21	0.01	0.06	0.06	44	0.24	0.01	0.07	0.07
123478-HxCDF	59	0.11	0.01	0.01	0.01	64	0.13	0.01	0.01	0.01	37	0.19	0.01	0.02	0.02
123678-HxCDF	77	0.17	0.01	0.02	0.02	85	0.16	0.01	0.02	0.02	47	0.21	0.01	0.02	0.02
123789-HxCDF	87	0.01	0.01	0.00	0.00	88	n.d.	0.01	0.00	n.d.	48	0.09	0.01	0.01	0.01
234678-HxCDF	68	0.23	0.01	0.02	0.02	74	0.19	0.01	0.02	0.02	40	0.32	0.01	0.03	0.03
1234678-HpCDF	71	0.14	0.01	0.00	0.00	79	0.13	0.01	0.00	0.00	44	0.27	0.02	0.00	0.00
1234789-HpCDF	81	0.02	0.02	0.00	0.00	84	0.01	0.02	0.00	0.00	42	0.12	0.02	0.00	0.00
OCDF	---	0.01	0.01	0.00	0.00	---	0.01	0	0.00	0.00	---	0.13	0.01	0.00	0.00
WHO (2005) - PCDDF/F TEQ excl. LOQ:					*0.60*					*0.54*					*0.74*
WHO (2005) - PCDDF/F TEQ incl. LOQ:				*0.60*					*0.54*					*0.74*	

Extraction method	**SER158**					**SER158**					**PLE**				
Sample intake g.	20.0055					20.0074					12.6419				
Moisture (%)	7.7					7.7					7.7				
Sample code:	SER158-3					SER158-4					PLE 3 +4 *1				
Compound Name	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ
2378-TCDD	86	0.09	0.01	0.09	0.09	84	0.09	0.01	0.09	0.09	47	0.10	0.01	0.10	0.10
12378-PeCDD	93	0.21	0.01	0.21	0.21	103	0.19	0.01	0.19	0.19	54	0.26	0.02	0.26	0.26
123478-HxCDD	73	0.05	0.01	0.01	0.01	69	0.03	0.01	0.00	0.00	44	0.04	0.02	0.00	0.00
123678-HxCDD	87	0.20	0.01	0.02	0.02	91	0.15	0.01	0.02	0.02	47	0.18	0.02	0.02	0.02
123789-HxCDD		0.02	0.01	0.00	0.00		0.03	0.01	0.00	0.00		0.02	0.02	0.00	0.00
1234678-HpCDD	87	0.12	0.01	0.00	0.00	85	0.10	0.01	0.00	0.00	50	0.18	0.02	0.00	0.00
OCDD	77	0.59	0.01	0.00	0.00	72	0.58	0.02	0.00	0.00	41	0.71	0.02	0.00	0.00
2378-TCDF	79	1.59	0.02	0.16	0.16	83	1.59	0.02	0.16	0.16	49	1.66	0.02	0.17	0.17
12378-PeCDF	85	0.39	0.01	0.01	0.01	88	0.37	0.01	0.01	0.01	51	0.44	0.02	0.01	0.01
23478-PeCDF	90	1.43	0.01	0.43	0.43	100	1.41	0.01	0.42	0.42	53	1.47	0.02	0.44	0.44
123478-HxCDF	69	0.16	0.01	0.02	0.02	66	0.16	0.01	0.02	0.02	40	0.15	0.02	0.01	0.01
123678-HxCDF	90	0.16	0.01	0.02	0.02	91	0.14	0.01	0.01	0.01	48	0.19	0.02	0.02	0.02
123789-HxCDF	94	0.00	0.01	0.00	0.00	92	0.00	0.01	0.00	0.00	50	0.01	0.02	0.00	0.00
234678-HxCDF	78	0.18	0.01	0.02	0.02	78	0.16	0.01	0.02	0.02	48	0.23	0.02	0.02	0.02
1234678-HpCDF	86	0.06	0.01	0.00	0.00	85	0.06	0.01	0.00	0.00	49	0.07	0.01	0.00	0.00
1234789-HpCDF	90	0.01	0.01	0.00	0.00	87	0.01	0.01	0.00	0.00	54	0.00	0.02	0.00	0.00
OCDF	---	0.01	0	0.00	0.00	---	0.01	0	0.00	0.00	---	0.01	0.01	0.00	0.00
WHO (2005) - PCDDF/F TEQ excl. LOQ:					*0.98*					*0.94*					*1.06*
WHO (2005) - PCDDF/F TEQ incl. LOQ:				*0.98*					*0.94*					*1.06*	

Extraction method	**SER158**					**SER158**					**PLE**				
Sample intake g.	20.0068					20.0062					19.2067				
Moisture (%)	6.5					6.5					6.5				
Sample code:	SER158-5					SER158-6					PLE 5 +6 *1				
Compound Name	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ
2378-TCDD	74	0.11	0.01	0.11	0.11	77	0.12	0.01	0.12	0.12	66	0.21	0	0.21	0.21
12378-PeCDD	81	0.24	0.01	0.24	0.24	86	0.28	0.01	0.28	0.28	76	0.35	0.01	0.35	0.35
123478-HxCDD	60	0.31	0.01	0.03	0.03	62	0.02	0.01	0.00	0.00	64	0.05	0.01	0.00	0.00
123678-HxCDD	83	0.18	0.01	0.02	0.02	83	0.22	0.01	0.02	0.02	66	0.29	0.01	0.03	0.03
123789-HxCDD		0.02	0.01	0.00	0.00		0.02	0.01	0.00	0.00		0.06	0.01	0.01	0.01
1234678-HpCDD	76	0.12	0.01	0.00	0.00	77	0.11	0.01	0.00	0.00	77	0.18	0.01	0.00	0.00
OCDD	65	0.13	0.01	0.00	0.00	64	0.15	0.01	0.00	0.00	69	0.29	0.01	0.00	0.00
2378-TCDF	72	1.92	0.01	0.19	0.19	75	2.09	0.01	0.21	0.21	65	2.93	0.01	0.29	0.29
12378-PeCDF	75	0.46	0.01	0.01	0.01	78	0.41	0.01	0.01	0.01	71	0.63	0.01	0.02	0.02
23478-PeCDF	78	1.73	0.01	0.52	0.52	81	1.89	0.01	0.57	0.57	73	2.14	0.01	0.64	0.64
123478-HxCDF	61	0.15	0.01	0.01	0.01	61	0.16	0.01	0.02	0.02	61	0.20	0.01	0.02	0.02
123678-HxCDF	80	0.18	0.01	0.02	0.02	82	0.17	0.01	0.02	0.02	72	0.26	0.01	0.03	0.03
123789-HxCDF	82	0.00	0.01	0.00	0.00	83	0.01	0.01	0.00	0.00	85	0.03	0.01	0.00	0.00
234678-HxCDF	70	0.20	0.01	0.02	0.02	70	0.19	0.01	0.02	0.02	65	0.28	0.01	0.03	0.03
1234678-HpCDF	77	0.09	0.01	0.00	0.00	76	0.06	0.01	0.00	0.00	76	0.10	0	0.00	0.00
1234789-HpCDF	78	0.01	0.02	0.00	0.00	77	0.01	0.01	0.00	0.00	79	0.04	0.01	0.00	0.00
OCDF	---	0.01	0.01	0.00	0.00	---	0.00	0.01	0.00	0.00	---	0.07	0	0.00	0.00
WHO (2005) - PCDDF/F TEQ excl. LOQ:					*1.18*					*1.27*					*1.64*
WHO (2005) - PCDDF/F TEQ incl. LOQ:				*1.18*					*1.27*					*1.64*	

*1 Results of PLE are combined

Continuation of Annex 1:

Detailed results fo six samples comparison of extraction SER158 versus PLE

DIOXINS

Extraction method	SER158					**SER158**					**PLE**				
Sample intake g.	20.01					20.0054					20.0582				
Moisture (%)	7.00					7.0					7.0				
Sample code:	SER158-1					SER158-2					PLE 1 +2 *1				
Compound Name	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ
PCB-77	58	14.7	0.03	0.002	0.002	56	14.2	0.02	0.001	0.001	23	14.4	0.05	0.001	0.001
PCB-81	71	0.4	0.02	0.000	0.000	66	0.5	0.02	0.000	0.000	25	0.7	0.05	0.000	0.000
PCB-105	75	734.2	0.02	0.022	0.022	73	682.7	0.01	0.021	0.021	38	677.9	0.02	0.020	0.020
PCB-114	78	48.1	0.02	0.001	0.001	71	45.8	0.01	0.001	0.001	37	47.3	0.02	0.001	0.001
PCB-118	84	2925.3	0.01	0.088	0.088	77	2700.2	0.01	0.081	0.081	39	2478.5	0.02	0.074	0.074
PCB-123	82	28.9	0.01	0.001	0.001	76	30.7	0.01	0.001	0.001	39	24.9	0.02	0.001	0.001
PCB-126	70	15.9	0.02	1.593	1.593	65	15.3	0.02	1.532	1.532	30	15.8	0.03	1.577	1.577
PCB-156	85	222.4	0.02	0.007	0.007	71	210.6	0.03	0.006	0.006	40	212.0	0.04	0.006	0.006
PCB-157	82	69.2	0.03	0.002	0.002	69	64.5	0.03	0.002	0.002	39	66.1	0.04	0.002	0.002
PCB-167	87	168.9	0.03	0.005	0.005	73	159.8	0.03	0.005	0.005	40	160.9	0.04	0.005	0.005
PCB-169	79	5.0	0.03	0.149	0.149	67	4.5	0.03	0.135	0.135	35	4.3	0.04	0.129	0.129
PCB-189	68	21.2	0.01	0.001	0.001	58	20.4	0.01	0.001	0.001	36	20.5	0.02	0.001	0.001
WHO (2005) - PCDDF/F TEQ excl. LOQ:					*1.870*					*1.786*					*1.818*
WHO (2005) - PCDDF/F TEQ incl. LOQ:				*1.870*					*1.786*					*1.818*	

Extraction method	**SER158**					**SER158**					**PLE**				
Sample intake g.	20.0055					20.0074					12.6419				
Moisture (%)	7.7					7.7					7.7				
Sample code:	SER158-3					SER158-4					PLE 3 +4 *1				
Entry identifier	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ
PCB-77	55	33.6	0.02	0.003	0.003	60	30.1	0.02	0.003	0.003	28	28.8	0.03	0.003	0.003
PCB-81	65	1.1	0.02	0.000	0.000	72	0.9	0.02	0.000	0.000	28	0.9	0.03	0.000	0.000
PCB-105	75	427.3	0.01	0.013	0.013	88	388.5	0.01	0.012	0.012	30	392.2	0.02	0.012	0.012
PCB-114	74	26.8	0.01	0.001	0.001	89	23.7	0.01	0.001	0.001	31	24.7	0.02	0.001	0.001
PCB-118	77	1377.6	0.01	0.041	0.041	92	1247.2	0.01	0.037	0.037	31	1177.6	0.02	0.035	0.035
PCB-123	77	15.8	0.01	0.001	0.001	92	15.7	0.01	0.001	0.001	31	14.6	0.02	0.000	0.000
PCB-126	65	11.7	0.02	1.165	1.165	72	10.5	0.01	1.053	1.053	27	10.1	0.03	1.014	1.014
PCB-156	76	161.7	0.02	0.005	0.005	89	145.2	0.02	0.004	0.004	31	150.7	0.03	0.005	0.005
PCB-157	73	42.2	0.02	0.001	0.001	87	37.5	0.02	0.001	0.001	30	38.8	0.03	0.001	0.001
PCB-167	80	94.4	0.02	0.003	0.003	93	84.9	0.02	0.003	0.003	32	87.2	0.03	0.003	0.003
PCB-169	68	2.2	0.02	0.066	0.066	79	2.0	0.03	0.060	0.060	30	2.1	0.03	0.063	0.063
PCB-189	61	16.1	0.01	0.001	0.001	71	14.9	0.01	0.000	0.000	26	14.8	0.02	0.000	0.000
WHO (2005) - PCDDF/F TEQ excl. LOQ:					*1.300*					*1.174*					*1.136*
WHO (2005) - PCDDF/F TEQ incl. LOQ:				*1.300*					*1.174*					*1.136*	

Extraction method	**SER158**					**SER158**					**PLE**				
Sample intake g.	20.0068					20.0062					19.2067				
Moisture (%)	6.5					6.5					6.5				
Sample code:	SER158-5					SER158-6					PLE 5 +6 *1				
Entry identifier	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ	Percent recovery 13C12 std	Calculated amount	Detection limit	WHO 2005 TEQ incl. LOQ	WHO 2005 TEQ excl. LOQ
PCB-77	55	37.6	0.02	0.004	0.004	57	35.6	0.03	0.004	0.004	58	39.6	0.03	0.004	0.004
PCB-81	64	0.8	0.02	0.000	0.000	67	0.8	0.02	0.000	0.000	72	0.8	0.02	0.000	0.000
PCB-105	71	637.5	0.01	0.019	0.019	78	592.1	0.01	0.018	0.018	85	656.6	0.02	0.020	0.020
PCB-114	70	36.3	0.02	0.001	0.001	78	34.2	0.01	0.001	0.001	87	36.2	0.02	0.001	0.001
PCB-118	73	2224.1	0.01	0.067	0.067	81	2125.3	0.01	0.064	0.064	90	2224.3	0.02	0.067	0.067
PCB-123	72	20.3	0.01	0.001	0.001	81	311.9	0.01	0.009	0.009	88	19.2	0.02	0.001	0.001
PCB-126	60	16.2	0.02	1.619	1.619	63	14.9	0.02	1.494	1.494	72	16.9	0.02	1.692	1.692
PCB-156	69	248.4	0.03	0.008	0.008	73	228.3	0.03	0.007	0.007	82	248.9	0.03	0.008	0.008
PCB-157	67	66.1	0.03	0.002	0.002	70	61.8	0.03	0.002	0.002	78	66.2	0.03	0.002	0.002
PCB-167	70	154.5	0.03	0.005	0.005	74	140.5	0.03	0.004	0.004	83	151.0	0.03	0.005	0.005
PCB-169	62	4.2	0.03	0.127	0.127	63	3.9	0.03	0.117	0.117	74	4.4	0.03	0.132	0.132
PCB-189	53	25.4	0.01	0.001	0.001	61	23.0	0.01	0.001	0.001	68	26.1	0.01	0.001	0.001
WHO (2005) - PCDDF/F TEQ excl. LOQ:					*1.852*					*1.721*					*1.931*
WHO (2005) - PCDDF/F TEQ incl. LOQ:				*1.852*					*1.721*					*1.931*	

*1 Results of PLE are combined