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RIVO Report

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The preparation of four biological reference materials for QUASIMEME

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Summary

Four biological materials have been prepared for use in QUASIMEME interlaboratory studies including a shrimp sample for metal analysis (QM01-1) and two mussel (QO01-3 and QO02-2) and one mackerel sample (QO02-1) for organic contaminant analysis.

1. Introduction

The QUASIMEME Project Office has requested RIVO to prepare 4 materials for the use in QUASIMEME interlaboratory studies (ILS) under contract QLTS/RIVO/1/02. The materials included a shrimp sample for metal analysis and two mussel and one mackerel sample for organic contaminant analysis.

2. Objectives

The objective of this work was the preparation of four homogeneous materials for the purpose of interlaboratory studies on organic contaminants and trace metals. The homogeneity of all materials should be tested and evaluated. In the materials a range of contaminants should be determined.

3. Materials and methods

3.1 Production of materials

The materials produced are mentioned in Table 1.

Table 1. Materials produced for QUASIMEME ILS.

No.	Code	Origin and purpose	No. of lots
1	QM01-1	Shrimps from the Dutch coast in glass jars for trace metal contaminant analysis with indicative analysis of As, Hg, Cd, Cr, Cu, Pb, Ni, Se and Zn.	250
2	QO01-3	Mussels from the German Bight in tins for organic contaminant analysis with indicative analysis for CBs, OCPs, PAHs, PBDEs, TBBP-A, meTBBP-A and HBCD.	250
3	QO02-1	Mackerel in tins, sampled north of Scotland for toxaphene analysis with indicative analysis for CBs, OCPs, toxaphene, PBDEs, TBBP-A, meTBBP-A and HBCD.	250
4	QO02-2	Mussels in tins from Loch Striven (UK) for organic contaminant analysis with indicative analysis for toxaphene, PCBs, OCPs, PAHs, PBDEs, TBBP-A, meTBBP-A and HBCD.	250

3.1.1 QM01-1 (Shrimps, Dutch coast)

Shrimps were caught by local fishermen at the Dutch coast close to IJmuiden. The shrimps have not been peeled but were cooked before further processing. Twenty-five kg of material was processed at 18 October 2001 as described below.

3.1.2 Q001-3 (Mussels, German Bight)

Ca 120 kg of fresh mussels originating from the German Bight have been purchased from a shellfish industry (Roem van Yerseke, Yerseke, Netherlands). The mussels were cleaned and transported to RIVO on 15 October 2001 where they were cooked for 5 min. and shells were removed. The meat was collected and frozen until further processing at 18 October 2001. Details on the processing are mentioned below.

3.1.3 Q002-1 (Mackerel, East of Shetlands)

Mackerel, originating from east of the Shetland Islands (61°00' N, 00°00' E/W) and caught by the Dutch trawler SCH 123, was purchased (round frozen) at 5 February 2002. After filleting it was frozen at -20°C for further processing. At 25 February 2002, the material was minced and the next day it was homogenised, tinned and sterilised as described below.

3.1.4 Q002-2 (Mussels, Loch Striven)

On 17 April 2003, 34 kg of mussel tissue has been received deep-frozen from dr. D Wells (personal delivery at RIVO). After thawing, it was found that the material contained a high moisture content, which would hamper the production of a homogeneous material. Therefore, it was decided to heat the raw material for a short period in order to reduce the moisture content. After heating, 11.75 kg of material remained, which was not sufficient for the production of 250 tins. The material was frozen at -20°C. At 4 June 2003, 15 kg cooked mussel tissue was received from the QUASIMEME project office, to mix with the earlier received material. At 12 June 2003, the material was thawed to room temperature, minced and frozen at -20°C. At 24 June 2003, the minced material was thawed, homogenised and tinned. Details on the production procedure are mentioned below.

3.1.5 Details on the procedure for production of materials

The complete volume of meat was minced using a mincer (Finis Machinefabriek, Ulft) in combination with a Fryma mill equipped with toothed rotary knives (Fryma Maschinen AG, Rheinfelden, Switzerland) to a final size of 3.5 mm². Subsequently, ca. 25 kg sample was homogenised for 3 minutes, after adding 0.02% butylhydroxytoluene (BHT), in a Stephan cutter (Stephan Machines, Almelo, The Netherlands), type UMM/SK25 (made in 1979). For the QO-coded materials, coated tins (Eurocan Food, Mechelen, Belgium, volume ca. 75 ml) were filled to the brim with homogenised material using a manual dosing machine (maschinenfabrik Engler, Vienna, Switzerland). The tins were sealed by a Lanico TVM 335 sealing machine (Thomassen and Drijver, Deventer, The Netherlands). For the QM-materials, glass jars were filled with ca 50 g of material and closed with the lid. The tins and jars were sterilised in a Muvero-Mat sterilizer (type 90E) for 45 minutes at 122 °C (pressure 1.4 bar, heating-time: 90 minutes, cooling time: 20 minutes).

Homogeneity was tested by analysing a representative contaminant in 10 selected lots in duplicate (ISO-Guide 35, 2001). For the QO coded materials (for organics), the homogeneity was tested by analysis of CB 118 and CB 153 (for QO01-3 and QO02-2). For QO02-1, the homogeneity was tested by analysis of CB 138 and CB 153, because CB118 was below the LOQ. The homogeneity of the QM coded materials was tested by analysis of Hg. For evaluation of the homogeneity, the Soft CRM software of BCR is used, which comprises ANOVA statistics.

3.2 Analytical determinations

The concentrations of CBs, OCPs and toxaphene were determined by gas chromatography and electron capture detection according to RIVO methods ISW A002 and ISW A011 (toxaphene). The samples were Soxhlet extracted with dichloromethane / n-pentane (1:1). The co-extracted fat was removed by alumina column chromatography. The PCBs were separated from OCPs or toxaphene by silica column chromatography prior to GC-ECD or GC-NCI-MS determination, respectively. Trans-nonachlor (TNC) was also determined by GC-NCI-MS. Calibration with technical toxaphene was used for the determination of total toxaphene, whereas the individual chlorobornanes are determined by calibration with the respective congeners.

The concentrations of BFRs were determined according to RIVO method ISW A102. The samples were Soxhlet extracted with hexane / acetone (3:1). After sulfuric acid treatment, co-extracted fat and contaminants were removed by gel permeation chromatography. The BFRs were separated from other contaminants by silica column chromatography and the collected fraction was treated with concentrated sulphuric acid prior to GC-NCI-MS determination.

The concentrations of PAHs were determined according to ISW A014. The fat in the samples was saponified by ethanolic potassium hydroxide and subsequently, the sample was extracted with n-hexane. After silica column chromatography clean-up, the final extract was analysed by HPLC-fluorescence detection.

The concentrations of zinc, copper, cadmium and lead were determined by inductively coupled plasma – mass spectrometry (ICP-MS) after microwave destruction at elevated pressure and temperature with 65% nitric acid (HNO_3), according to RIVO method ISW A099.

The determination of the total mercury concentration (the anorganic and organic fraction) was carried out according to ISW A021. The samples were microwave destructed with 10% nitric acid (HNO_3) and subsequently the total mercury concentration was determined by flow injection analysis and flameless atomic absorption spectrometry.

Selenium was determined according to ISW A020. Samples were microwave destructed with 70% nitric acid (HNO_3) and 30% hydrogenperoxide (H_2O_2) and the destruate is reduced by 37% hydrogen chloride (HCl). The hydride of selenium was formed by addition of sodium boron hydride in order to facilitate the measurement with flow injection analysis and atomic absorption spectrometry.

Arsenic was determined according to the RIVO method ISW A047. The sample was incinerated and ashes were dissolved and after addition of a reagent, the arsenic complex was spectrophotometrically determined.

The determination of chromium and nickel was carried out by TNO-Nutrition, Zeist, The Netherlands. The fat content was determined according to RIVO method ISW A004 and is an adjusted chloroform/methanol extraction based on the method of Bligh and Dyer .

The moisture content was determined gravimetrically by drying the sample for 3 hours at 105°C, according to RIVO method ISW A034.

The determinations are accredited under ISO17025 lab no. L097 and the quality is assured by internal and external reference materials, blank and recovery tests and by the participation in interlaboratory studies.

4. Results and discussion

The homogeneity testing results for each material are discussed in paragraph 4.1. The results of the indicative analysis are discussed in paragraph 4.2.

4.1 Results of the homogeneity tests

4.1.1 Results of material QM01-1 (shrimps, Dutch coast)

The results of the homogeneity study of material QM01-1 are shown in Table 2.

Table 2. Homogeneity test results of material QM01-1 for mercury (mg/kg ww).

Sample ID	Analysis #1	Analysis #2	Mean	STDev		Repeated analysis
05	0.023	0.022	0.023	0.000		0.025 0.026
30	0.021	0.018	0.020	0.002		0.023
55	0.02	0.022	0.021	0.001		0.028
80	0.019	0.019	0.019	0.000		0.026
105	0.022	0.024	0.023	0.001		0.025
130	0.027	0.026	0.027	0.000		0.025
155	0.026	0.027	0.027	0.001		0.025
180	0.028	0.026	0.027	0.001		0.025
205	0.026	0.027	0.027	0.001		0.024
230	0.027	0.025	0.026	0.001		0.025 0.025
		Mean	0.024		Mean	0.025
					Stdev	0.001
					CV(%)	4.7

ANOVA Table

Source of Variation	SS*	d.f.*	MS*	StDev	F	F-crit 95%	F-crit 99%
Between Units	17.7*10 ⁻⁵	9	1.97*10 ⁻⁵	0.003	13.582	3.020	4.942
Within Units	1.45*10 ⁻⁵	10	1.45*10 ⁻⁶	0.001			
Total	19.2*10 ⁻⁵	19					

* SS: sum of squares, d.f.: degrees of freedom, MS: mean squares

Snedecor F-Test

Differences between units statistically significant? (α=95%) :Yes

Differences between units statistically significant? (α=99%) :Yes

Due to a miscommunication, jars 5 to 105 were analysed in a separate sequence from jars 130 to 230. Due to a small blank contribution in the first sequence in combination with the very low mercury levels in the sample, the levels (after correction for the blank) are lower compared to the second sequence samples. This negatively influences the ANOVA statistics suggesting a between unit inhomogeneity. However, each sample has been analysed again ('repeated analysis'). The data from 'Repeated analysis' (Table 2) show a good RSD of 4,7%, indicating a good between unit homogeneity. Sample ID 5 and 230 were analysed in duplicate, confirming

the good within unit homogeneity that was already shown in the initial homogeneity analysis (by comparison of the analysis 1 & 2 values of each lot).

The within unit standard deviation ranges from 0 (due to rounding) to 0.002 and the between unit standard deviation is in the same range (0.001 and 0.002). The between unit coefficient of variation in the repeated analysis was 4.7-9.9% which is lower compared with results generally observed in interlaboratory studies. In round 32 of the QUASIMEME ILS, for flounder sample QTM057BT between lab CVs at low levels ($\mu\text{g}/\text{kg}$ range) ranged from 15.1% for mercury to 123% for silver (QUASIMEME, 2003).

4.1.2 Results on material Q001-3 (mussels, German Bight)

The homogeneity results of material Q001-3 are shown in Tables 3 and 4. The graphs of the distribution of the means of the duplicate analysis are shown in Appendix 1.

Table 3. Homogeneity results of material Q001-3 for CB 118 ($\mu\text{g}/\text{kg}$ ww).

Sample ID	Analysis #1	Analysis #2	Mean	STDev
5	1.79	1.84	1.815	0.035
30	1.87	2.07	1.970	0.141
55	1.96	1.86	1.910	0.071
80	1.71	1.78	1.745	0.049
105	1.82	1.84	1.830	0.014
130	1.86	1.95	1.905	0.064
155	1.85	1.78	1.815	0.049
180	1.86	1.75	1.805	0.078
205	1.88	1.76	1.820	0.085
230	1.81	1.75	1.780	0.042
		Mean	1.840	
		STDev	0.068	
		CV(%)	3.70	
		n	20	

ANOVA Table

Source of Variation	SS	d.f.	MS	StDev	F	F-crit 95%	F-crit 99%
Between Units	0.083	9	0.009	0.046	1.833	3.020	4.942
Within Units	0.050	10	0.005	0.071			
Total	0.134	19					

Snedecor F-Test

Differences between units statistically significant? ($\alpha=95\%$): No

Differences between units statistically significant? ($\alpha=99\%$): No

Table 4. Homogeneity results of material Q001-3 for CB 153 ($\mu\text{g}/\text{kg ww}$).

Sample ID	Analysis #1	Analysis #2	Mean	STDev
5	6.21	6.26	6.235	0.035
30	6.42	6.45	6.435	0.021
55	6.47	6.24	6.355	0.163
80	5.93	6.09	6.010	0.113
105	6.1	6.13	6.115	0.021
130	6.08	6.46	6.270	0.269
155	6.22	6.27	6.245	0.035
180	6.28	6.12	6.200	0.113
205	6.31	6.11	6.210	0.141
230	6.21	6.22	6.215	0.007
		Mean	6.229	
		STDev	0.117	
		CV(%)	1.87	
		n	20	

ANOVA Table

Source of Variation	SS	d.f.	MS	StDev	F	F-crit 95%	F-crit 99%
Between Units	0.245	9	0.027	0.079	1.845	3.020	4.942
Within Units	0.148	10	0.015	0.122			
Total	0.393	19					

Snedecor F-Test

Differences between units statistically significant? ($\alpha=95\%$):NoDifferences between units statistically significant? ($\alpha=99\%$):No

From the data of both CB 118 and CB 153 it can be seen that at both the 95 and 99% confidence levels the variance within a tin is in the same range as between tins. As is shown by the F-test, the material can be considered as homogeneous, both within and between tins. The CVs (3.7 and 1.9% for CB 118 and CB 153, respectively) are very low and do not contribute significantly to the between lab CVs in an ILS. In round 32 of the QUASIMEME ILS, for mussel sample QOR074BT between lab CVs ranged from 18.7% for CB 153 to 105% for o,p-DDT (QUASIMEME, 2003).

4.1.3 Material Q002-1 (mackerel, East of Shetlands)

The homogeneity results of material Q002-1 are shown in Table 5 and Table 6.

Table 5. Homogeneity results of material Q002-1 for CB 138 ($\mu\text{g}/\text{kg ww}$).

Sample ID	Analysis #1	Analysis #2	Mean	STDev
5	1.50	1.39	1.45	0.08
55	1.36	1.30	1.33	0.04
80	1.30	1.26	1.28	0.03
105	1.42	1.29	1.36	0.09
130	1.34	1.34	1.34	0.00
155	1.34	1.54	1.44	0.14
205	1.23	1.41	1.32	0.13
230	1.49	1.56	1.53	0.05
255	1.33	1.49	1.41	0.11
285	1.48	1.29	1.39	0.13
		Mean	1.38	
		STDev	0.07	
		CV(%)	5.28	
		n	20	

ANOVA Table

Source of Variation	SS	d.f.	MS	StDev	F	F-crit 95%	F-crit 99%
Between Units	0.096	9	0.011	0.032	1.232	3.020	4.942
Within Units	0.087	10	0.009	0.093			
Total	0.183	19					

Snedecor F-Test:

Differences between units statistically significant? ($\alpha=95\%$): No

Differences between units statistically significant? ($\alpha=99\%$): No

Table 6. Homogeneity results of material Q002-1 for CB 153 ($\mu\text{g}/\text{kg ww}$).

Sample ID	Analysis #1	Analysis #2	Mean	STDev
5	2.79 (outlier)	1.69	2.24	0.78
55	1.88	2.01	1.95	0.09
80	1.73	1.52	1.63	0.15
105	1.80	1.74	1.77	0.04
130	1.84	1.85	1.85	0.00
155	1.78	1.76	1.77	0.01
205	1.65	1.89	1.77	0.17
230	1.88	2.11	2.00	0.16
255	1.74	1.92	1.83	0.13
285	2.16	1.87	2.02	0.21
		General statistical data (without outlying tin 5)		
		Mean	1.84	
		STDev	0.17	
		CV(%)	9.22	
		n	18	

ANOVA Table

Source of Variation	SS	d.f.	MS	StDev	F	F-crit 95%	F-crit 99%
Between Units	0.253	8	0.032	0.088	1.952	3.230	5.467
Within Units	0.146	9	0.016	0.127			
Total	0.399	17					

Snedecor F-Test

Differences between units statistically significant? (a=95%) :No

Differences between units statistically significant? (a=99%) :No

Tin 5 showed an outlier for CB 153 (not for CB 138) and was therefore excluded from ANOVA statistics for this CB. The ANOVA results for CB 153 showed a somewhat higher standard deviation for both the within and between results compared with CB 138. From the data of both CB 138 and CB 153 it can be seen that at 95 and 99% confidence level the variance within a tin is in the same range as between tins. Therefore, the material can be considered as homogeneous, both within and between tins, although the variance is somewhat higher than normally observed (e.g. materials Q001-3 and Q002-2). The CVs (5.3 and 9.2% for CB 138 and CB 153, respectively) are low compared to the between lab CVs in an ILS. In round 32 of the QUASIMEME ILS, for herring sample QOR075BT between lab CVs ranged from 18.5% for CB 153 to 131% for p/p-DDT (QUASIMEME, 2003).

4.1.4 Results of material Q002-2 (mussels, Loch Striven)

The homogeneity test results of material Q002-2 are shown in Tables 7 and 8.

The graphs on the distribution of the means of the duplicate analysis are shown in Appendix 1.

Table 7. Homogeneity results of material Q002-2 for CB 118 ($\mu\text{g}/\text{kg ww}$).

Sample ID	Analysis #1	Analysis #2	Mean	STDev
5	0.93	1.00	0.965	0.049
30	0.93	1.02	0.975	0.064
55	0.94	0.97	0.955	0.021
80	1.01	0.97	0.990	0.028
105	0.95	1.02	0.985	0.049
130	0.98	0.95	0.965	0.021
155	0.91	0.95	0.930	0.028
180	1.02	0.97	0.995	0.035
205	0.98	1.00	0.990	0.014
230	0.96	0.99	0.975	0.021
	General statistical data			
		Mean	0.973	
		STDev	0.02	
		CV(%)	2.031	
		n	20	

ANOVA Table

Source of Variation	SS	d.f.	MS	StDev	F	F-crit 95%	F-crit 99%
Between Units	0.012	9	0.001	MSB < MSW	0.667	3.020	4.942
Within Units	0.020	10	0.002	0.045			
Total	0.032	19					

Snedecor F-Test:

Differences between units statistically significant? (a=95%) :No

Differences between units statistically significant? (a=99%) :No

Table 8. Homogeneity results of material Q002-2 for CB 153 (µg/kg ww).

Sample ID	Analysis #1	Analysis #2	Mean	STDev
5	2.77	2.85	2.810	0.057
30	2.70	3.03	2.865	0.233
55	2.76	2.70	2.730	0.042
80	2.9	2.79	2.845	0.078
105	2.68	2.81	2.745	0.092
130	2.76	2.72	2.740	0.028
155	2.81	2.74	2.775	0.049
180	2.78	2.71	2.745	0.049
205	2.60	2.77	2.685	0.120
230	2.74	2.76	2.750	0.014
		General statistical data		
		Mean	2.769	
		STDev	0.055	
		CV(%)	2.001	
		n	20	

ANOVA Table

Source of Variation	SS	d.f.	MS	StDev	F	F-crit 95%	F-crit 99%
Between Units	0.042	9	0.005	MSB < MSW	0.467	3.020	4.942
Within Units	0.100	10	0.010	0.100			
Total	0.142	19					

Snedecor F-Test

Differences between units statistically significant? (a=95%) :No

Differences between units statistically significant? (a=99%) :No

From the data of both CB 118 and CB 153 it can be seen that at the 95 and 99% confidence level the variance within a tin is in the same range as that between tins. Therefore, the material can be considered as homogeneous, both within and between tins. The CVs (2.0 and 2.0% for CB 118 and CB 153, respectively) are very low do not contribute significantly to the between lab CVs in an ILS. In round 32 of the QUASIMEME ILS, for mussel sample QOR074BT between lab CVs ranged from 18.7% for CB 153 to 105% for o'p-DDT (QUASIMEME, 2003).

4.2 Results of the indicative analysis of contaminants

The results of the indicative analysis of QM01-1 are shown in Table 9. Except for lead, all trace metals in the sample are at levels above the limit of quantification (LOQ). Therefore, this material can be used for trace metal interlaboratory studies.

Table 9. Indicative levels of trace metals in shrimp (QM01-1)

Determinand	Concentration (mg/kg ww)
As	1.59
Hg	0.025
Cd	0.052
Cr	0.10
Cu	10.1
Pb	<0.05
Ni	0.25
Se	0.54
Zn	23
Lipid (g/kg)	22
Moisture (%)	74.2

The results of the indicative analysis of Q001-3, Q002-1 and Q002-2 are shown in table 10. In material Q001-3, the levels of some OCPs (HCHs, HCB, transnonachlor o,p'- and p,p'-DDT) are close or below LOQ of the method used, whereas p,p'-DDD, p,p'-DDE and dieldrin are at levels above LOQ. Moreover, half of the BFR compounds in material Q001-3 (BDE 28, 66, 71, 85, 119, 153, 154, 183 and 190) exhibit concentration levels below LOQ. The congeners predominantly observed in marine and freshwater fish samples (BDE congeners 47, 99 and 100) are just above the LOQ. Therefore, this material might be less suitable for an OCP or BFR interlaboratory study that covers all OCPs and BFRs, although laboratories can obviously also report their <LOQ values to QUASIMEME. Nearly all PAHs are above the LOQ. All over it is concluded that this material is very suitable for (PCB- and PAH-) interlaboratory studies. The level of only one of the PAH compounds is below the LOQ.

The levels of several PCBs in the materials Q002-1 (CB 28, 52, 101, 105, 118, 156 and 180), several OCPs (?-HCH, transnonachlor, o,p'-, p,p'-DDT and p,p'-DDD) and nine of the BFR compounds are below the LOQ of the method used, whereas the other contaminants are at levels above LOQ. This material will be very useful to evaluate laboratories performance at levels at or close to their LOQs. Detectable levels of toxaphene make this sample also suitable for toxaphene interlaboratory studies.

For material Q002-2, the levels of some OCPs (?-?-HCH, transnonachlor and p,p'-DDT) are below the LOQ of the method used in the material (?-HCH was not determined), whereas the other contaminants are at quantifiable levels. The recovery of o,p'-DDT is 53% and is therefore considered to be unreliable. Furthermore, the levels of several BFRs are below the LOQ, but the major BFR congeners (47, 99 and 100) are at levels above their LOQ. Three out of fifteen PAHs are below the LOQ of the method used. Levels of all toxaphene congeners are below the LOQ. It is concluded that material Q002-2 will be very useful to perform (PCB- and PAH-) interlaboratory studies and that the material might also be suitable to evaluate laboratories performances at levels close to their LOQs, regarding OCP and BFR contaminants.

Table 10. Indicative levels of organic contaminants in the materials

Group	Determinant	Concentration (µg/kg ww)		
		Q001-3	Q002-1	Q002-2
		Mussel (German Bight)	Mackerel	Mussel (Loch Striven)
PCBs	CB-28	0.2	<0.8	0.3
	CB-52	0.5	<0.7	0.6
	CB-101	2	<2	1.3
	CB-105	-	<0.8	0.4
	CB-118	1.6	<2	0.9
	CB-153	5.6	1.8	2.8
	CB-138	3.5	1.4	2.1
	CB-156	-	<0.8	0.2
	CB-180	0.3	<0.9	0.5
	OCPs	Dieldrin	1.2	4.9
HCB		<0.1	1.8	0.1
?-HCH		<0.1	0.8	<0.06
?-HCH		<0.2	<1	<0.3
?-HCH		<0.1	nd*	nd
o'-p-DDT		<0.2	<2	0.6**
p'-p-DDE		1.3	4.3	3.6
p'-p-DDD		0.5	<0.8	1.6
p'-p-DDT		<0.2	<2	<0.4
Transnonachlor		<0.1	<1	<0.2
BFRs	BDE 28	<0.1	<0.1	<0.2
	BDE 47	0.4	1.0	2.8
	BDE 66	<0.1	0.2	0.2
	BDE 71	<0.1	<0.1	<0.1
	BDE 75	0.3	<0.1	<0.1
	BDE 77	0.2	<0.1	<0.1
	BDE 85	<0.1	<0.1	<0.1
	BDE 99	0.1	0.4	1.7
	BDE 100	0.1	0.3	0.6
	BDE 119	<0.1	<0.1	<0.1
	BDE 138	0.1	<0.1	<0.1
	BDE 153	<0.1	<0.1	nd
	BDE 154	<0.1	0.1	<0.1
	BDE 190	<0.1	<0.1	<0.1
	HBCD	<0.1	<0.3	<0.1
	TBBPA	nd	nd	nd
	PAHs	Acenaftene	0.9	-
Fluorene		2.7	-	2
Fenantrene		16	-	5.9
Anthracene		0.7	-	0.2
Fluorantene		15	-	3.2
Pyrene		8.9	-	2.1
Benzo(a)anthracene		1.9	-	4.4
Chrysene		3.0	-	<0.2
Benzo(e)pyrene		4.7	-	4
Benzo(b)fluorantene		3.3	-	1.9
Benzo(k)fluorantene		1.4	-	0.5
Benzo(a)pyrene		1.0	-	<0.3
Dibenz(a,h)anthracene		<1	-	0.5

Table 10 (continued)

	Benzo(g,h,i)perylene	1.9	-	0.9
	Indeno(1,2,3-cd)pyrene	2.0	-	1.5
Toxaphene	CHB-26	-	1.3	<0.3
	CHB-50	-	2.5	<0.4
	CHB-62	-	1.3	<0.3
	Total toxaphene	-	50	<41
Lipid (%)		2.8	24.8	5.2
Moisture (%)		nd	58.8	72.3

*nd: not determined

** Not accurate due to low recovery (53%)

5. Conclusions

Four biological reference materials (QM01-1, Q001-3, Q002-1 and Q002-2) of 250 tins each have successfully been prepared. The between- and within homogeneity were satisfactory, although a somewhat higher variance was observed in sample Q002-1 for unknown reasons. The variances as observed in these homogeneity tests were all much lower compared to values normally observed in interlaboratory studies.

Material QM01-1 is very suitable for a metal interlaboratory study. Material Q001-3 is suitable for a PCB and PAH interlaboratory study. Material Q002-1 is very suitable for a OCP and toxaphene ILS and Q002-2 can be used for nearly all contaminant groups (excluding toxaphene). Although some contaminant groups of material Q001-3 and Q002-1 are close or below the LOQ, they represent samples that are analysed in everyday routine and can therefore be used to evaluate laboratories performance at these very low levels.

6. References

- ISO guide 35 (2001). Certification of reference materials - general and statistical principles, 3rd ed., draft 2. Geneva, International Organization for Standardization.
- QUASIMEME (2003). QUASIMEME laboratory performance studies, BT-1 Trace metals in biota, round 32 - exercise 549, QUASIMEME, Aberdeen.
- QUASIMEME (2003). QUASIMEME laboratory performance studies, BT-2 Chlorobiphenyls and organochlorine pesticides in biota, round 32 - exercise 550, QAUSIMEME, Aberdeen.

Acknowledgement

Marco Lohman is gratefully acknowledged for the statistical analysis of the homogeneity results and the preparation of the graphs.

Appendix 1. Homogeneity graphs (distribution of the means of duplicate analysis)

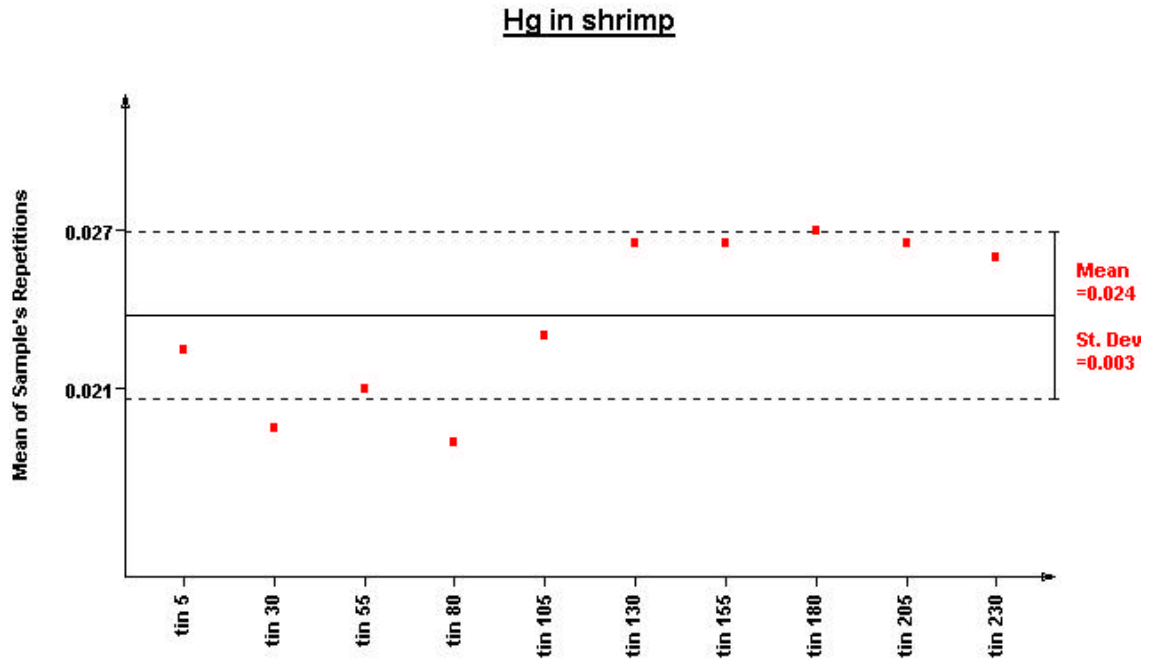


Figure 1. Means of two replicate analysis of each jar the homogeneity study for mercury in material QM01-1

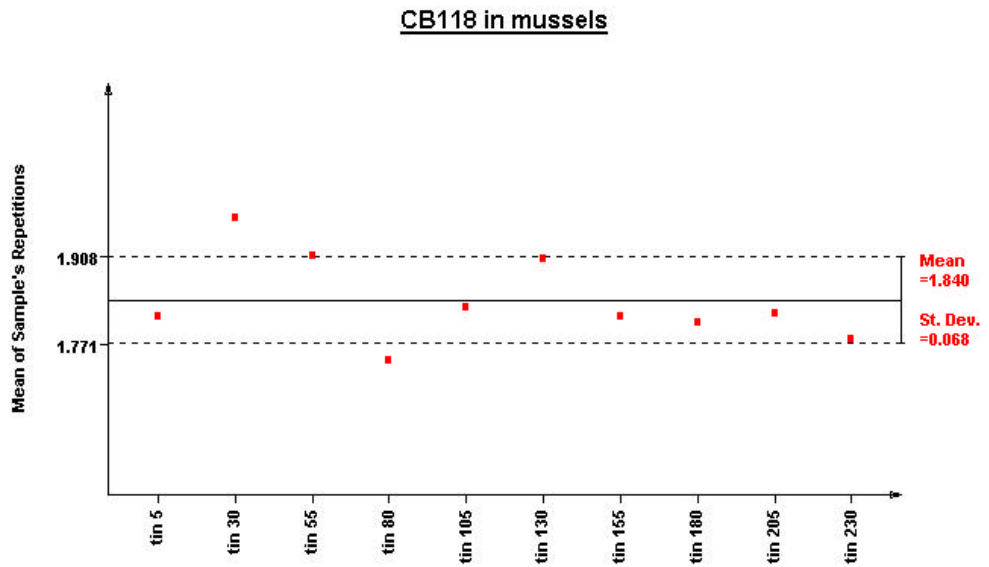


Figure 2. Means of two replicate analysis of each tin in the homogeneity study for CB 118 in material Q001-3

CB153 in mussels

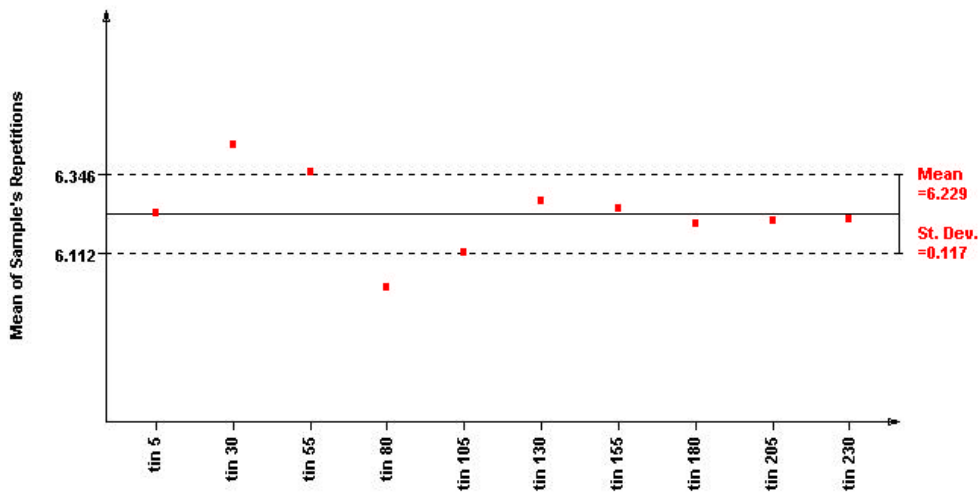


Figure 3. Means of two replicate analysis of each tin in the homogeneity study for CB 153 in material Q001-3

Graph for CB138

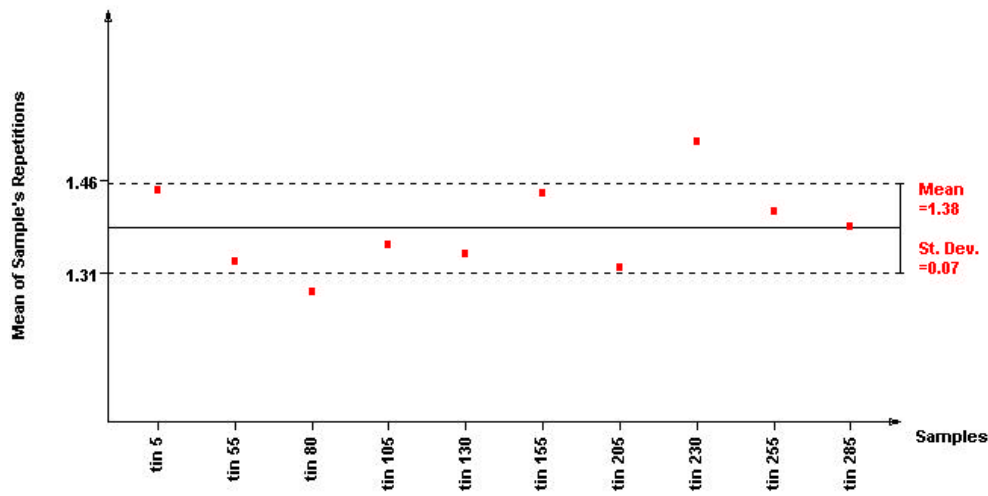


Figure 4. Means of two replicate analysis of each tin in the homogeneity study for CB 138 in material Q002-1

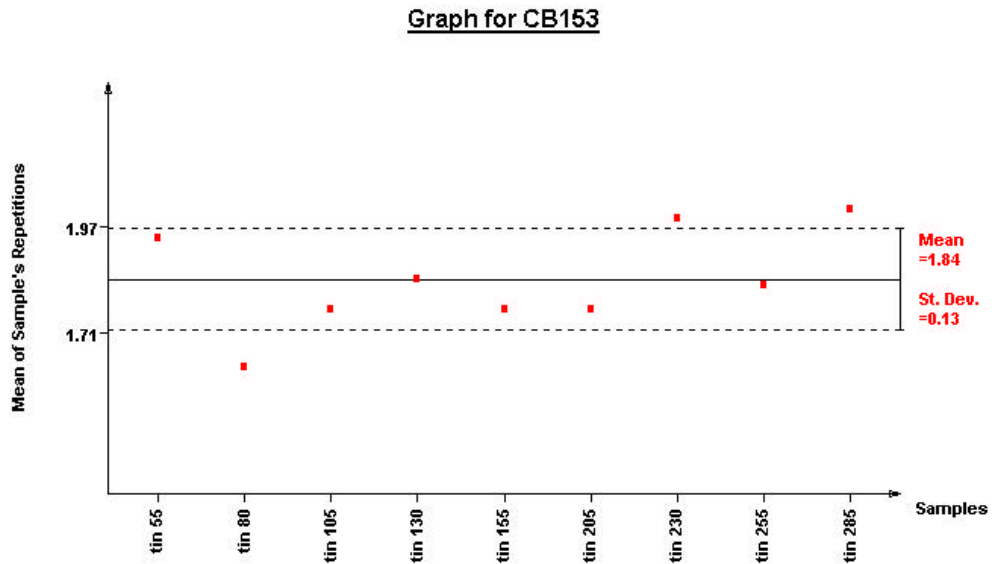


Figure 5. Means of two replicate analysis of each tin in the homogeneity study for CB 153 in material Q002-1. Tin 5 is excluded (outlier).

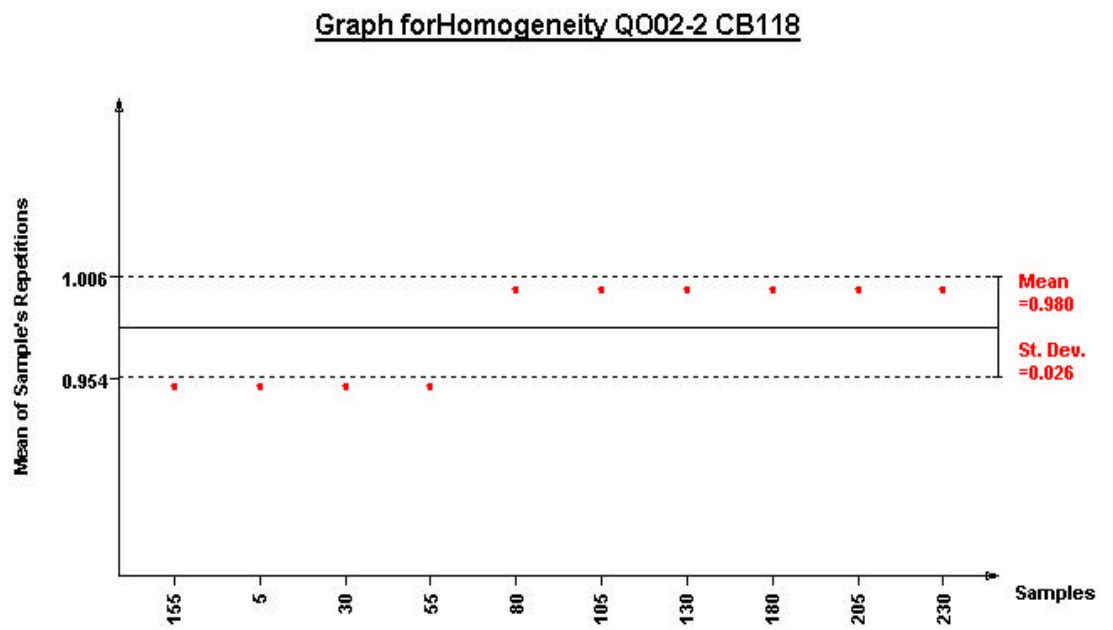


Figure 6. Means of two replicate analysis of each tin in the homogeneity study for CB 118 in material Q002-2

Graph for Homogeneity Q002-2 CB153

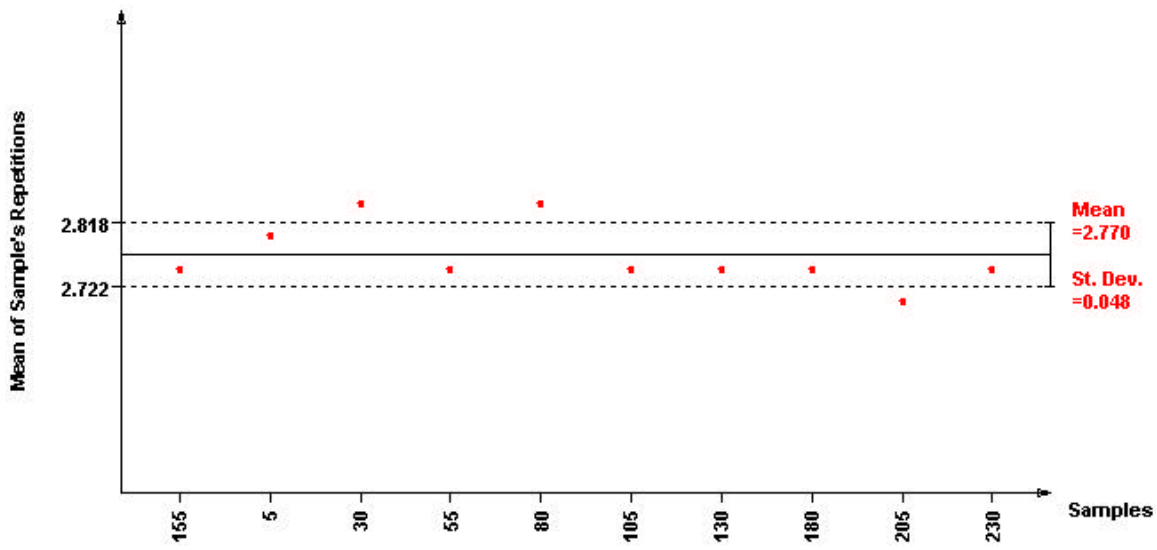


Figure 7. Means of two replicate analysis of each tin in the homogeneity study for CB 153 in material Q002-2.