D3.2 INTER-CALIBRATION STUDY RESULTS

VERSION 1.0

Martin Söderström[1], Vesa Häkkinen[1] and Olli Kostiainen[1]

Helsinki 28.09.2012

[1] Finnish Institute for Verification of the Chemical Weapons Convention (VERIFIN)

Baltic Sea Region Programme 2007-2013

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1. Introduction

1.1 General

Transnational inter-calibration was organized to study the selected methods for analysis of certain sea-dumped chemical weapons and/or their degradation products.

The inter-calibration was coordinated by VERIFIN (PP6).Three other partners – IO PAS (PP1), MUT (PP3) and FOI (PP4) received samples for analysis and also VERIFIN analysed the samples. MUT, FOI and VERIFIN analysed the samples for organic chemical weapons and/or their degradation products and IO PAS analysed the samples for organic and total arsenic.

The aim of the study was to assess transferability of the methods between laboratories and variability of the results of different laboratories. Additionally, the idea was to test the selected analysis method.

Based on the study the partners plan to build and maintain an international laboratory cooperation scheme, which aims to exchange of samples, experience and reference materials.

1.2 Conduct of the study

The inter-calibration study was discussed in the CHEMSEA project meeting in Stockholm 29.2.–2.3.2012. Based on the discussions, VERIFIN proceeded with the preparation of the study.

The stability of the spiked samples were tested over a 20-day period. After confirmation of the satisfactory stability of the planned chemicals in the used matrix, the preparation of the samples started.

Samples were prepared for three laboratories performing analysis of organic chemicals and for one laboratory performing arsenic measurements. The samples were shipped on 25.5.2012.

Reports were received from all participants (18.6.–27.6.2012 for organic analysis and 3.8 for arsenic measurements) and the study results were evaluated by VERIFIN. The preliminary results were presented and discussed in the CHEMSEA project meeting in Helsinki 12.–14.9.2012.

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2. Chemicals

Based on preliminary stability tests done by VERIFIN, the chemicals presented in Table 1 were selected for the stability test.

The two first chemicals (thiodiglycol and 1,4-dithiane) are degradation products of sulphur mustard. Triphenylarsine is a component in arsine oil and triphenylarsine oxide is its oxidation product.

Table 1. Chemicals selected for the inter-calibration study.

Chemical name	CAS Number	Structure	Source	Purity
Thiodiglycol	111-48-8		Aldrich	≥99%
1,4-Dithiane	505-29-3		Aldrich	≥97%
Triphenylarsine	603-32-7		Acros Organics	≥97%
Triphenylarsine oxide	1153-05-5		Aldrich	≥97%
Arsine in nitric acid (2–3%)	7440-38-2	As	Merck	CertiPUR

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3. Samples

3.1 Stability test

Prior to sample dispatch the stability of the test chemicals in the corresponding sediment matrix was checked for a period of 20 days. Selected concentrations are shown in Table 2 below.

Table 2. The selected concentrations of chemicals for the stability study.

Chemical name	Amount	
	Low concentration	High concentration
Thiodiglycol	667 ng/g	6670 ng/g
1,4-Dithiane	667 ng/g	6670 ng/g
Triphenylarsine	166 ng/g	1660 ng/g
Triphenylarsine oxide	166 ng/g	1660 ng/g

Preparation of the samples

A 3.0-g portion of dry sediment was placed into 10 ml Falcon tubes. 100 µl of acetone solution containing 20 µg of 1,4-dithiane, 5.0 µg triphenylarsine, 20 µg of thiodiglycol and 5.0 µg of triphenylarsine oxide was spiked to the high concentration sediment samples and ten times lower concentration to the low concentration sediment samples. The closed Falcon tubes were shaken for 5 minutes and kept overnight in a room temperature. 7 ml of distilled water containing 0.1 % dichloromethane was added to the sample tubes and the tubes were kept overnight in a refrigerator to ensure the moistening of the sediment. The next day the sample tubes were placed into freezer (-20 °C).

Sample preparation

Sample preparation scheme for GC–MS samples and LC–MS/MS samples is presented in Figure 1. Three parallel samples were analysed after 2, 12 and 20 days from spiking.

The samples were first centrifuged for 5 minutes at 2000 G. The excess pore water was collected. Quality blank was prepared with the samples using an empty 50 ml Falcon tube.

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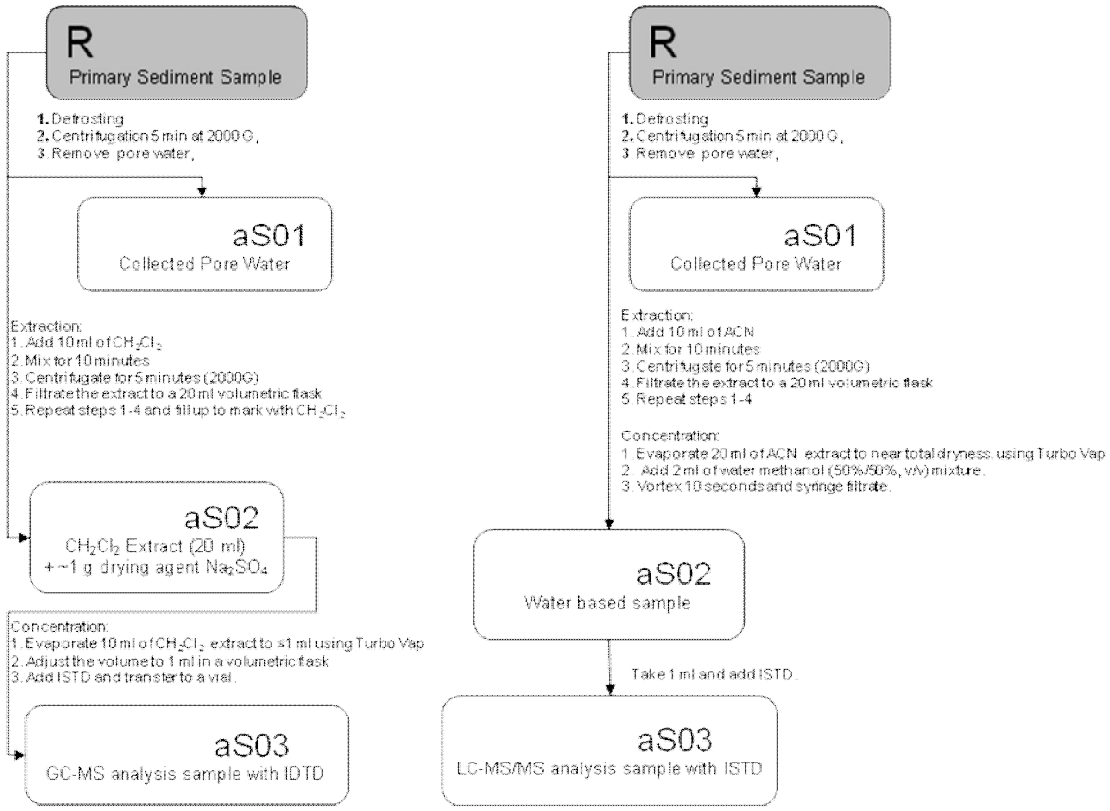

Figure 1. Sample preparation scheme used at VERIFIN for the stability test.

Samples for GC–MS

The samples were extracted with 10 ml of dichloromethane (Fluka, PESTANAL stabilized with 25 mg/l amylene) by shaking for 10 minutes (1500 rpm). The samples were centrifuged for 5 minutes at 2000 G and the dichloromethane layer was decanted trough filter paper (Whatman 1 PS, rinsed with CH_2Cl_2) into a 20 ml volumetric flask. The extraction was repeated with another 10 ml portion of dichloromethane, the dichloromethane layers from the two extractions were combined and the 20 ml volumetric flask was filled to the volume with dichloromethane by rinsing the filter paper. The extract was transferred into an 20 ml EPA vial and approximately 1 g of anhydrous sodium sulphate (Merck, Suprapur) was added. The organic extract was left to stand overnight in a refrigerator.

A 10 ml of fraction, of the extract was transferred into an evaporation vessel and concentrated below 1 ml using mild nitrogen flow and heating (5 psi, 23 °C with Turbo Vap). The concentrated sample was adjusted to a volume of 1 ml in a volumetric flask by dichloromethane. Exact amount of approximately 200 ppb HCB internal standard solution (same amount as in quantitation standards) was added to the sample. The sample was transferred into an autosampler vial for analysis.

Samples for LC–MS/MS

The samples were extracted with 10 ml of acetonitrile (VWR, HiPerSolv) by shaking for 10 minutes (1500 rpm). The samples were centrifuged for 5 minutes at 2000 G and the acetonitrile layer was decanted trough filter paper (Whatman, qualitative 4) into a 20 ml volumetric flask. The extraction was repeated with another 10 ml portion of acetonitrile and the acetonitrile layers from the two extractions were combined. The extract was transferred into an evaporation vessel and evaporated to near dryness using mild nitrogen flow and heating (7.5 psi, 44 °C with Turbo Vap). The evaporation residue was dissolved by adding 2 ml of a mixture of purified water and LC quality methanol (50%/50%, v/v) and vortexing for 10 seconds. The sample was filtered using a syringe filter (Millex PTFE/LCR 0.45µm) into a glass (?) vial. An exact 1 ml fraction of the filtrate was transferred into an autosampler (?) vial and an exact amount of 200 ppb DMMP solution (same amount as in quantitation standards) was added.

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GC–EI/MS method

All samples were analysed using an Agilent 6890N gas chromatograph (GC) equipped with an automatic liquid injector and an Agilent 5975B mass selective detector (MSD). Capillary column was a DB-5ms (Agilent, 30 m x 0.25 mm i.d., 0.25 µm film). The column temperature was programmed from 40 °C (isothermal time 1 min) to 290 °C at 10 °C/min and held at final temperature for 10 min. Splitless injection (splitless time 1 min) was used with the injector temperature of 250 °C. The carrier gas was helium with a flow of 35 cm/sec at 40 °C. The transfer line between the GC and MSD temperature was kept at 290 °C. The ionisation mode was electron impact (electron energy 70 eV), with an electron multiplier voltage of 300 V above the "autotune" value for lower concentration samples and "autotune" value for higher concentration samples. Selected ion monitoring (SIM) with three selective ions was used for identification and quantitation. The ions used for SIM are presented in Table 3. A six-point calibration (ca. 10–350 pg/µl) was applied for lower concentration samples and (ca. 100–3500 pg/µl) for higher concentration samples. Hexachlorobenzene (HCB) was used as internal standard (ISTD).

Table 3. Ions used for SIM for the GC–EI/MS analysis.

Chemical	SIM ions (m/z)	
	Quantifier	Qualifier
1,4-Dithiane	61	120, 92
Triphenylarsine	306	227, 152
HCB (ISTD)	288	286, 284

LC–APCI/MS/MS method

The LC–MS/MS analysis was performed using a ThermoScientific Accela liquid chromatograph and ThermoScientific TSQ Quantum Ultra triple quadrupole mass spectrometer. The analysis was done using atmospheric pressure chemical ionisation (APCI) technique followed by selective reaction monitoring (SRM) (see Table 4 for conditions) using argon as collision gas. All compounds were analyzed in positive ion mode using 7 µA discharge current, 250 °C capillary temperature and 450 °C vaporizer temperature. A six-point calibration (ca. 10–450 pg/µl) was applied for lower concentration samples and (ca. 250–6000 pg/µl) for higher concentration samples. Dimethyl methylphosphonate (DMMP) was used as internal standard (ISTD).

Table 4. The SRM conditions used for the LC–MS/MS analysis.

Chemical	Reaction (*m/z*)		Collision energy (eV)
	Quantifier	Qualifier	
Thiodiglycol	105 > 87	105 > 61 105 > 41	20
Triphenylarsine oxide	323 > 227	323 > 154	33, 38
DMMP (ISTD)	125 > 93	125 > 111	20

Results of the stability study

Table 5 presents the results obtained from the stability test for a period to 20 days. Based on these results it can be stated that no degradation of spiked chemicals was observed. The recoveries of triphenylarsine after 20 days was as high as 132 % at low concentration sample whereas the recoveries at high concentration sample was between 73.7 % to 79.6 %. One reason for this can be the poor peak shape observed especially at low concentration level making the quantitation difficult.

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Table 5. Results of stability study

Spiking chemical	Technique	Spiked (ng/g)	Day 0			Day 10			Day 20		
			Measured (ng/g)	Average	Recovery (%)	Measured (ng/g)	Average	Recovery (%)	Measured (ng/g)	Average	Recovery (%)
Thiodiglycol	LC–API/MS/MS	667	232 203 222	219	32.9	201 194 201	199	29.8	145 227 190	188	28.1
		6670	2820 2920 2840	2870	43.0	2650 2820	2730	41.0	3310 3050 3950	3430	51.5
1,4-Dithiane	GC–EI/MS	667	50.4 45.7 46.0	47.4	7.1	56.6 49.9 52.4	53.0	7.9	56.0 72.7 59.1	62.6	9.4
		6670	1180 970 1220	1130	16.9	1180 1050 1240	1160	17.3	1440 1360 1250	1350	20.2
Triphenylarsine	GC–EI/MS	166	211 145 175	177	106	168 172	170	102	201 230 226	219	132
		1660	1350 1170 1440	1320	79.6	1180 1260	1220	73.7	1210 1310 1310	1280	76.8
Triphenylarsine oxide	LC–API/MS/MS	166	196 184 196	192	116	185 177 186	183	110	167 165 155	162	98
		1660	1730 1600 1710	1680	101	1760 1780	1770	107	1900 1850 2060	1930	116

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3.2 Preparation of the samples

Based on stability study, the final adjustments to the concentration of spiking chemicals were made (See Table 6).

Table 6. The selected concentrations of chemicals for the inter-calibration study.

Chemical name	Amount	
	Low concentration	High concentration
Thiodiglycol	269 ng/g	2690 ng/g
1,4-Dithiane	667 ng/g	6670 ng/g
Triphenylarsine	170 ng/g	1700 ng/g
Triphenylarsine oxide	66.7 ng/g	667 ng/g
Arsine	75 µg/g	200 µg/g

Organic samples

3.0 g of dry sediment was placed into 10 ml Falcon tubes. 100 µl of acetone solution containing 20 µg of 1,4-Dithiane, 5,1 µg triphenylarsine, 8,06 µg of thiodiglycol and 2,0 µg of triphenylarsine oxide was spiked to the high concentration sediment samples and ten times lower concentration to the low concentration sediment samples.

The closed Falcon tubes were shaken for 5 minutes and kept overnight in the room temperature. 7ml of distilled water containing 0.1 % dichloromethane was added to the sample tubes and kept overnight in the refrigerator to ensure the moistening of the sediment. The next day the sample tubes were placed into freezer (temperature?).

Inorganic samples

3.0 g of dry sediment was placed into 10 ml Falcon tubes. 0.225 ml of water solution containing 0.225 mg and 0.6 ml of water solution containing 0.6 mg arsine were spiked to the low and high concentration samples, respectively. The closed Falcon tubes were shacked for 5 minutes and kept overnight in the room temperature. 7ml of distilled water containing 0.1 % dichloromethane was added to the sample tubes and kept overnight in the refrigerator to ensure the moistening of the sediment. In the next day the sample tubes were placed into freezer.

Quality control standards

Quality control standard solutions of HCB for GC–MS (50 µg/ml and 50 ng/ml in dichloromethane) and DMMP for LC–MS (50 µg/ml and 50 µg/ml in water) were prepared.

HCB solution (50 µg/ml) was placed into 2 ml glass ampoules, which were first frozen with liquid nitrogen and then sealed by melting with propane/air flame. DMMP and low concentration HCB (?) solutions were placed into screw-capped Agilent autosampler vials, which were closed tightly and wrapped in parafilm.

3.3 Packaging and transport of the samples

All samples were spiked in 50 ml Falcon plastic tubes (primary containers) which were closed tightly with plastic stopper. The primary containers were packed in polyethylene zip-lock bags and placed inside the plastic secondary containers. The empty space in the secondary container was filled with vermiculite and the container was closed with a sealable lid.

All primary and secondary sample containers were labelled as described in the attachments placed in each sample package before closing and sending to participating laboratories. The secondary containers were placed inside a polystyrene box. Before closing the boxes, plates of dry ice were inserted into each

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box to keep the samples cool during the sample transport from VERIFIN to the participating laboratories.

Each polystyrene box was placed inside a cardboard box which was filled to the top with plastic packing material. Each cardboard box was tightly closed with adhesive tape and labelled according to TNT Express instructions before sample dispatch. Samples were delivered to participating laboratories by TNT Express and the sample dispatch took place on 25 May 2012.

Participating laboratories were notified the planned sample dispatch date (25 May 2012) by e-mail on 22 May 2012.

The following pages show the letter sent with the samples.

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Accompanying letter to laboratories A, B and C

HELSINGIN YLIOPISTO
HELSINGFORS UNIVERSITET
UNIVERSITY OF HELSINKI

VERIFIN

CHEMSEA
CHEMICAL MUNITIONS
SEARCH & ASSESSMENT

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Attachment

1(1)

24.5.2012 VER-VH-0196

Finnish Institute for Verification of
the Chemical Weapons Convention

Ref:	CHEMSEA
Title:	**Attachment to Round-Robin samples**
Keywords:	CHEMSEA, Sample analysis, Round-Robin
Distribution:	Participating laboratories

Dear Sir/Madam,

Please find attached samples for the CHEMSEA Round-Robin test.

Two sets of sediment samples are delivered, one set for GC-based analysis and another set for LC-based analysis. Samples are coded as follows:

X/GC1, X/GC2, X/GC3, X/LC1, X/LC2, and X/LC3, where

X = participating laboratory (A, B, or C)

GC = samples for GC-based analysis

LC = samples for LC-based analysis

Each sample set contains two samples and one blank sample.

Standard solutions of Hexachlorobenzene for GC (HCB, 50 ppm and 50 ppb solutions in dichloromethane) and Dimethylmethyl phosphonate for LC (DMMP, 50 ppm and 50 ppb solutions in water) are delivered with the samples.

VERIFIN will send separate instructions for sample analysis to each participating laboratory, and **laboratories must not start analysing the samples before the instructions have been received!**

Samples should be stored in a freezer below -18 ºC until the sample analysis is started.

Kemiallisen aseen kieltosopimuksen instituutti (VERIFIN) PL 55 (A.I. Virtasen aukio 1), 00014 Helsingin yliopisto
Puhelin (09) 191 50443 (toimisto), faksi (09) 191 50437, www.verifin.helsinki.fi

Verifikationinstitutionen för konventionen mot kemiska vapen (VERIFIN) PB 55 (A.I. Virtanens plats 1), FIN-00014 Helsingfors universitet
Telefon +358 9 191 50443, fax +358 9 191 50437, www.verifin.helsinki.fi

Finnish Institute for Verification of the Chemical Weapons Convention (VERIFIN) P.O. Box 55 (A.I. Virtasen aukio 1), FIN-00014 University of Helsinki
Telephone +358 9 191 50443, fax +358 9 191 50437, www.verifin.helsinki.fi

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Accompanying letter to laboratory D

HELSINGIN YLIOPISTO
HELSINGFORS UNIVERSITET
UNIVERSITY OF HELSINKI

CHEMSEA
CHEMICAL MUNITIONS SEARCH & ASSESSMENT

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VERIFIN

Attachment

1(1)

24.5.2012 VER-VH-0197

Finnish Institute for Verification of
the Chemical Weapons Convention

Ref:	CHEMSEA
Title:	**Attachment to Round-Robin samples**
Keywords:	CHEMSEA, Sample analysis, Round-Robin
Distribution:	Participating laboratories

Dear Sir/Madam,

Please find attached samples for the CHEMSEA Round-Robin test.

Sediment samples containing arsene are delivered. Samples are coded as follows:

X/As1, X/As2, and X/As3, where

X = participating laboratory (D or E)
As = sample containing arsene

One of the samples is a blank sample.

Samples should be stored in a freezer below -18 ºC until the sample analysis is started.

Kemiallisen aseen kieltosopimuksen instituutti (VERIFIN) PL 55 (A.I. Virtasen aukio 1), 00014 Helsingin yliopisto
Puhelin (09) 191 50443 (toimisto), faksi (09) 191 50437, www.verifin.helsinki.fi

Verifikationinstitutionen för konventionen mot kemiska vapen (VERIFIN) PB 55 (A.I. Virtanens plats 1), FIN-00014 Helsingfors universitet
Telefon +358 9 191 50443, fax +358 9 191 50437, www.verifin.helsinki.fi

Finnish Institute for Verification of the Chemical Weapons Convention (VERIFIN) P.O. Box 55 (A.I. Virtasen aukio 1), FIN-00014 University of Helsinki
Telephone +358 9 191 50443, fax +358 9 191 50437, www.verifin.helsinki.fi

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4. Results of the analysis inter-calibration study

4.1 Analysis of organic chemicals

The results of the analysis are summarised in Tables 7 and 8. The first table present the actual reported concentrations and the second table presents the recoveries.

It should be noted that these recoveries are **not** real recoveries as they are a combination of two factors:

- part of the chemical is removed with the pore water which is centrifuged and removed prior to the analysis; in real samples the chemicals are typically bound strongly to the sediment and not easily removed with the water phase,
- part of the chemical is bound to the sediment (*this is the normal recovery*).

Results from laboratories A and B are quite similar, but laboratory C had some problems with the analysis. These problems were discussed in the Helsinki meeting and further analysis will be performed by laboratory C after resolving issues related to the analysis.

The sample preparation methods are summarised in Table 9 and the mass spectrometric methods in Table 10.

For the reporting of the inter-calibration study, the laboratories were requested to provide ion or transition ratios, which are often used for confirmation of the identification. Table 11 summarises the recorded ratios. Both laboratories A and B obtained satisfactory ratios for verified identification. Laboratory C will remeasure the ratios during further analysis.

Thiodiglycol findings

The findings between labs A and B differ to a degree, but these difference might be explained by different conditions of the samples during transport. As the samples to laboratory B melted en route, it could be possible that more thiodiglycol was transferred to the water phase that in the sample for laboratory A.

1,4-Dithiane findings

Both analysis methods and the findings of all three laboratories were quite similar. The higher recovery of 1,4-dithiane by laboratory C might be explained similar contamination as with the blank sample.

The reason relatively low recovery of 1,4-dithiane of ca. 20 % is not clear, but some experiments will be carried out at VERIFIN e.g. with different types of concentration procedures.

Triphenylarsine and triphenylarsine oxide findings

It is noticeable that the recoveries of triphenylarsine oxide are clearly above 100 %. Between laboratories A and B there is a trend: A has higher triphenylarsine recoveryand B had higher triphenylarsine oxide recovery.

This could be explained by the effect of sample transport and the conditions of the sample. The sample to Laboratory A was not shipped, but was just stored at freezer until the sample preparation started. The samples to laboratory B were shipped frozen and were received melted.

The figure 2 clearly shows that the amount of triphenylarsine has diminished in the samples for laboratory B probably due spontaneous oxidation to triphenylarsine oxide as well as possibly via binding to the matrix. At the same time the amount of triphenylarsine oxide has increased through oxidation of triphenylarsine leading to recoveries exceeding 100 %.

Laboratory C reported the following observation in their report:

> "As a result of preliminary tests, we found that chromatographic analysis of triphenylarsine, carried out in recommended conditions, may contribute an incorrect results, because the injector temperature was too high, because triphenylarsine decomposes above 220 ˚C, and as a result peaks are too low area, and also peaks strongly tailing, which may affect the accurate quantification, especially at low concentrations.
>
> We think that triphenylarsine oxide can be analyzed by gas chromatography, although its molecules lose an oxygen atom in injector by thermal decomposition and goes into the chromatography column as triphenylarsine. Therefore, determination triphenylarsine oxide and

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triphenylarsine in the same sample using gas chromatograph may fraught with a large error, or may even unenforceable."

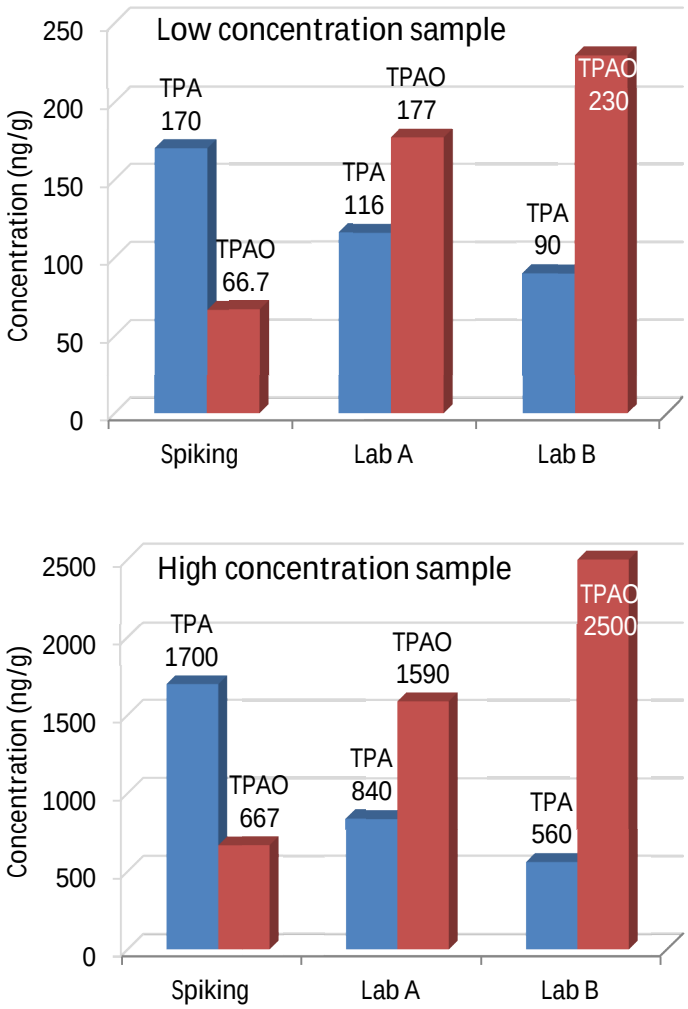

Figure 2. Spiking concentrations (in ng/g) as well findings by laboratories A and B for triphenylarsine (TPA) and triphenylarsine oxide (TPAO).

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Table 7. Analysis results of the inter-calibration study. Amounts are given in ng/g of sediment.

Spiking chemical	Method	Blank Sample				Low concentration sample				High concentration sample			
		Spiking	Lab A	Lab B	Lab C	Spiking	Lab A	Lab B	Lab C	Spiking	Lab A	Lab B	Lab C
Thiodiglycol	GC–MS	0	*n.a.*	–	19726	269	*n.a.*	15	10513	2690	*n.a.*	65	12902
	LC–MS		–	*n.a.*	19167		120	83	2833		1350	710	6267
1,4-Dithiane	GC–MS	0	–	–	151	667	100	100	246	6670	1350	1500	1569
Triphenylarsine	GC–MS	0	–	–	90	170	116	90	284	1700	840	560	1399
Triphenylarsine oxide	GC–MS	0	*n.a.*	*n.a.*	< 17	66.7	*n.a.*	*n.a.*	< 17	667	*n.a.*	*n.a.*	2138
	LC–MS		–	–	1984		177	230	594		1590	2500	1433

– Nothing found (but analysed)
n.a. Not analysed

Table 8. Analysis results of the inter-calibration study. Amounts are given in % of the spiking amount.

Spiking chemical	Method	Low concentration sample			High concentration sample		
		Lab A	Lab B	Lab C	Lab A	Lab B	Lab C
Thiodiglycol	GC–MS	*n.a.*	6 %	3908 %	*n.a.*	2 %	480 %
	LC–MS	45 %	31 %	1053 %	50 %	26 %	233 %
1,4-Dithiane	GC–MS	15 %	15 %	37 %	20 %	22 %	24 %
Triphenylarsine	GC–MS	68 %	53 %	167 %	49 %	33 %	82 %
Triphenylarsine oxide	GC–MS	*n.a.*	*n.a.*	10 %	*n.a.*	*n.a.*	321 %
	LC–MS	104 %	135 %	349 %	238 %	375 %	215 %

– Nothing found (but analysed)
n.a. Not analysed

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Table 9. Summary of sample preparation methods

Analysis	Laboratory A	Laboratory B	Laboratory C
GC–MS analysis	**A-1** Centrifugation, removal of water Extraction using 20 ml dichloromethane Shaking, centrifugation, filtration, drying Concentration Addition of ISTD (HCB) *Overall concentration factor: 10:1*	**B-1** Centrifugation, removal of water Extraction using 20 ml dichloromethane Drying Concentration Addition of ISTD Derivatisation using BSTFA *Overall concentration factor: 10:1*	**C-1** Centrifugation, removal of water Extraction using 25 ml dichloromethane Centrifugation, filtration, drying Concentration Addition of ISTD (HCB) *Overall concentration factor: 5:1*
		B-2 Centrifugation, removal of water Extraction using 20 ml dichloromethane Drying Concentration Addition of ISTD Concentration *Overall concentration factor: 50:1*	**C-2** Centrifugation, removal of water Extraction using 25 ml acetonitrile Shaking, centrifugation, filtration Concentration Derivatisation using TFAI, shaking Addition of ISTD (HCB) *Overall concentration factor: 1:1*
LC–MS analysis	**A-2** Centrifugation, removal of water Extraction using 20 ml acetonitrile Shaking, centrifugation, filtration, drying Evaporation to near dryness Dissolution in water/methanol (50:50) Addition of ISTD (D) *Overall concentration factor: 10:1*	**B-3** Centrifugation, removal of water Extraction using 20 ml acetonitrile Evaporation to dryness, solvent addition Addition of ISTD (DMMP) *Overall concentration factor: 10:1*	**C-3** Centrifugation, removal of water Extraction using 25 ml acetonitrile Shaking, centrifugation, filtration Evaporation to near dryness Dissolution in water/methanol (50:50) Addition of ISTD (DMMP) *Overall concentration factor: 5:1*
		B-4 Centrifugation, removal of water Extraction using 20 ml acetonitrile Evaporation to dryness, solvent addition Addition of ISTD (DMMP) Evaporation to dryness Removal of residual water Derivatisation using BSTFA *Overall concentration factor: 10:1*	

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Table 10. Summary of analysis methods

Chemical	Method	Laboratory A	Laboratory B	Laboratory C
Thiodiglycol (TDG)	GC–MS		**B-2** *As BSTFA derivative* GC–EI/MS (Q) SIM: *m/z* 116 *(Quantifier)* *m/z* 147 *(not used)* *m/z* 176 **B-4** *As BSTFA derivative* GC–EI/MS (Q) SIM: *m/z* 116 *(Quantifier)* *m/z* 147 *(not used)* *m/z* 176	**C-2** *As TFAI derivative* GC–EI/MS/MS (QQQ) SRM: *m/z* 141 → 113 *(Quantifier)* *m/z* 141 → 69 Collision: 12.5 eV w/ N_2
	LC–MS	**A-2** *As original chemical* LC–APCI+/MS/MS (QQQ) SRM: *m/z* 105 → 87 *(Quantifier)* *m/z* 105 → 61 *m/z* 105 → 41 Collision: 20 V w/ Ar	**B-3** *As original chemical* LC–ESI+/MS (QQQ) SIM: *m/z* 105 (also *m/z* 139 for TDG sulfoxide)	**C-3** *As original chemical* LC–APCI+/MS/MS (QQQ) SRM: *m/z* 105 → 61 *(Quantifier)* *m/z* 105 → 87 Collision: 9 V w/ N_2
1,4-Dithiane (DT)	GC–MS	**A-1** *As original chemical* GC–EI/MS (Q) SIM: *m/z* 61 *(Quantifier)* *m/z* 92 *m/z* 120	**B-1** *As original chemical* GC–EI/MS (Q) SIM: *m/z* 120 *(Quantifier)* *m/z* 94 *m/z* 61 *(not used)*	**C-1** *As original chemical* GC–EI/MS/MS (QQQ) SRM: *m/z* 120 → 105 *(Quantifier)* *m/z* 120 → 61 Collision: 15 eV using N_2
Triphenylarsine (TPA)	GC–MS	**A-1** *As original chemical* GC–EI/MS (Q) SIM: *m/z* 306 *(Quantifier)* *m/z* 277 *m/z* 152	**B-1** *As original chemical* GC–EI/MS (Q) SIM: *m/z* 152 *(Quantifier)* *m/z* 227 *(not used)* *m/z* 306	**C-1** *As original chemical* GC–EI/MS/MS (QQQ) SRM: *m/z* 306 → 152 *(Quantifier)* *m/z* 306 → 306 Collision: 10.6 eV w/ N_2
Triphenylarsíne oxide (TPAO)	GC–MS			**C-2** *As TPA (see text)* GC–EI/MS/MS (QQQ) SRM: *m/z* 306 → 152 *(Quantifier)* *m/z* 306 → 306 Collision: 10.6 eV w/ N_2
	LC–MS	**A-2** *As original chemical* LC–APCI+/MS/MS (QQQ) SRM: *m/z* 323 → 227 *(Quantifier)* *m/z* 323 → 154 Collision: 33 & 38 V w/ Ar	**B-3** *As original chemical* LC–ESI+/MS (QQQ) SIM: *m/z* 323	**C-3** *As original chemical* LC–APCI+/MS/MS (QQQ) SRM: *m/z* 323 → 227 *(Quantifier)* *m/z* 323 → 154 Collision: 37 & 29 V w/ N_2

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Table 11. Summary of calculated ion ratios supporting the identifications: calculated ratio (ratio from reference material).

Chemical	Method	Derivative	Blank sample			Low concentration sample			High concentration sample		
			Lab A	Lab B	Lab C	Lab A	Lab B	Lab C	Lab A	Lab B	Lab C
Thiodiglycol	GC–MS	BSTFA	*n.a.*	*n.a.*	–	*n.a.*	12 % (13 %)	–	–	15 % (14 %)	–
			n.a.	*n.a.*	–	*n.a.*	15 % (14 %)	–	–	13 % (14 %)	–
		TFAI	*n.a.*	*n.a.*	??	*n.a.*	*n.a.*	??	–	–	??
	LC–MS	original	–	–	??	64 % (63 %)	*n.d.*	??	61 % (62 %)	*n.d.*	??
1,4-Dithiane	GC–MS	original	–	–	??	44 % (47 %)	57 % (59 %)	??	46 % (47 %)	59 % (60 %)	??
Triphenylarsine	GC–MS	original	–	–	??	28 % (23 %)	28 % (28 %)	??	25 % (26 %)	24 % (25 %)	??
Triphenylarsíne oxide	GC–MS	original	*n.a.*	*n.a.*	??	–	–	??	–	–	??
	LC–MS	original	–	–	??	60 % (61 %)	x	??	60 % (60 %)	x	??

– Nothing found (but analysed)
n.a. Not analysed
x Only one ion/transition
?? Not available at the moments

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4.2 Analysis of arsenic

Only IO PAS (PP1; Laboratory D) analysed the arsenic samples as the other laboratory planning to take part in the arsenic measurements could not arrange the conduct of the analysis in time.

Two different analysis were done by laboratory D: total arsenic measurement and inorganic arsenic measurement. The reported organic arsenic amount are the difference of the two measurements. The results are summarised in Table 12 below.

The values of the inorganic arsenic measurements are very close tot he spiking values. The organic arsenic values, however, do not reflect the amount of organic arsenic present in the sample.

Table 12. Summary of arsenic determinations

Sample	Laboratory D			Spiking		
	Inorganic As	Organic As	Total	Inorganic As	Organic As	Total
Blank sample	10	1.0	11	–	–	–
Low concentration sample	73	1.1	74	75	1.2	76
High concentration sample	192	1.1	193	200	12	212

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5. Conclusion

The inter-calibration study was conducted according to the original plan.

All three laboratories planned to conduct the analysis of organic chemicals participated the study. Only one of the two laboratories planned fort he arsenic measurements could participate.

The results of the analysis were mostly satisfactory, but some issues have to be solved before the analysis of actual samples can take place. An updated version of this document will be produced to incorporate all the actions taken.

The behaviour of the chemicals during the study have also to be discussed to find the best approach for the sample handling and preparation.

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