# Quasimeme Laboratory Performance Studies


## Round 2014 - 2

1 October 2014 to 1 January 2015

Exercise Protocols

Version 1: 28th August 2014

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## Introduction Round 2014 - 2

Thank you for participating in the 2014 QUASIMEME Laboratory Performance studies.

The test materials for the exercises in Round 2014-2, that you have ordered will be sent to you by courier in the week of 6 October 2014. Please check that the contents of your package are correct and that all test materials are intact. If any test materials have been damaged in transit or if the wrong samples have been send, use the form in Annex 1 of this document to request replacement materials within two weeks after receipt of the test materials.

Additional test materials may also be purchased from QUASIMEME.

This protocol covers the following studies :

| Round | Analysis Group Code | Matrix | Analytes |
|---|---|---|---|
| 2014 - 2 | AQ-1 | Seawater | Nutrients |
| 2014 - 2 | AQ-2 | Estuarine and Low Salinity Open Water | Nutrients |
| 2014 - 2 | AQ-3 | Seawater | Metals |
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| 2014 – 2 | AQ-12 | Seawater | Organotins |
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| 2014 - 2 | AQ-14 | Seawater | DOC |
| 2014 - 2 | MS-1 | Sediment | Trace Metals |
| 2014 - 2 | MS-2 | Sediment | Chlorinated Organics |
| 2014 - 2 | MS-3 | Sediment | Polycyclic Aromatic Hydrocarbons |
| 2014 - 2 | MS-6 | Sediment | Organotins |
| 2014 - 2 | MS-7 | Sediment | Brominated Flame Retardants |
| 2014 - 2 | BT-1 | Fish or Shellfish | Trace Metals |
| 2014 - 2 | BT-2 | Fish or Shellfish | Chlorinated Organics |
| 2014 - 2 | BT-4 | Shellfish | Polycyclic Aromatic Hydrocarbons |
| 2014 - 2 | BT-8 | Biota | Organotins |
| 2014 - 2 | BT-9 | Fish or Shellfish | Brominated Flame Retardants |
| 2014 - 2 | BT-7 | Shellfish and Solution | ASP Shellfish Toxins |
| 2014 - 2 | BT-11 | Shellfish and Solution | DSP Shellfish Toxins |
| 2014 – 2 | BT-12 | Shellfish | PSP Shellfish Toxins |

All data for these studies must be uploaded to your Quasimeme Participant site, using the data submission forms, no later than 1 January 2015

IMPORTANT: Please note that the data submission forms are changed, report ONLY the parameters as they are asked in this protocol

All other information should be sent to: QUASIMEME Project Office

| | |
|---|---|
| QUASIMEME Project Office<br>Wageningen UR<br>PO Box 8005<br>NL-6700 EC Wageningen<br>The Netherlands | Website: http://www.Quasimeme.org<br>Tel.: +31 (0) 317 48 65 46<br>Fax: +31 (0) 317 485666<br>E-mail: Quasimeme@wur.nl |

| ROUND | 2014 - 2 |
|---|---|
| AQ-1 Nutrients in Seawater | |
| Test materials | SAMPLE 1, SAMPLE 2 and SAMPLE 3 |

**Objective**

This study covers the determination of nutrients in the seawater test materials.

**Test Materials and storage**

The test materials were prepared at the MUMM Laboratory, Ostend, Belgium, using seawater collected from the Atlantic Ocean aboard of the R.V. Belgica.

The seawater was filtered using a 0.45µm / 0.2µm double membrane filter. The pH of the filtered seawater was adjusted to ~ pH 7.2 with 0.1M hydrochloric acid and spiked to appropriate concentrations. The spiked seawater is then thoroughly mixed before being dispensed into the glass and plastic bottles. The filled bottles are then autoclaved at 110°C, 1.5 bar for 30 minutes. The autoclaving process removes micro-organisms, which affect the stability of the nutrient test materials. It has been demonstrated that autoclaving the test materials generates an increase in pH. We have found that after autoclaving, the pH of the nutrient test materials is within the range pH 7.5 to 8.5.

The three test materials differ from each other in respect of their nutrient concentrations.

There are two bottles for each test material - one glass and one plastic. The glass bottle should only be used for the determination of TOxN, nitrite, ammonia and total-N. The plastic bottle should only be used for the determination of silicate, phosphate and total-P. Each bottle contains approximately 250 ml of the test material.

Each batch of material was prepared in bulk. Homogeneity testing is performed on each batch of test materials produced. The nutrient test materials are stable for the period of the test, and have also been shown to be stable for a period of some months, even after opening, if used under the correct conditions.

Test materials should be stored in a refrigerator at +4°C, and analysed as soon as possible after receipt. Once the test materials are opened they should be analysed immediately.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Seawater (Salinity > 30 psu) |
| SAMPLE 2 | Seawater (Salinity > 30 psu) spiked |
| SAMPLE 3 | Seawater (Salinity > 30 psu) spiked |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

### Determinands and concentration ranges

The following nutrients should be determined:

| Determinand | Unit | Concentration range | | Error | | AA-EQS |
|---|---|---|---|---|---|---|
| | | Seawater | Seawater (spiked) | Const | Prop | |
| Ammonia | µmol/L | 0.2—5 | 0.2—5 | 0.1 | 6.0% | |
| Nitrite | µmol/L | 0.01—2 | 0.01—2 | 0.01 | 6.0% | |
| Phosphate | µmol/L | 0.05—5 | 0.05—5 | 0.05 | 6.0% | |
| Silicate | µmol/L | 0.5—10 | 0.5—10 | 0.1 | 6.0% | |
| Total-N | µmol/L | 5—25 | 5—25 | 0.5 | 6.0% | |
| Total-P | µmol/L | 0.1—5 | 0.1—5 | 0.05 | 6.0% | |
| TOxN | µmol/L | 0.05—15 | 0.05—15 | 0.05 | 6.0% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

### Analysis

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material "as received". The concentration of nutrients should be determined against your own calibration solutions.

### Reporting

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 -2 |
|---|---|
| AQ-2 Nutrients in Estuarine and low salinity open seawater | |
| Test materials | SAMPLE 1, SAMPLE 2, SAMPLE 3 and SAMPLE 4 |

**Objective**

This study covers the determination of nutrients in estuarine water test materials and low salinity open water test materials.

**Test Materials and storage**

The test materials were prepared at the MUMM Laboratory, Ostend, Belgium, using seawater collected from the Atlantic Ocean (Estuarine water samples) and from the Baltic Sea (low salinity water samples)

The seawater was filtered using a 0.45µm / 0.2µm double-membrane filter and diluted with ultrapure demineralised water. The pH of the filtered seawater was adjusted to ~pH7.2 with 0.1M hydrochloric acid and spiked to appropriate concentrations. The spiked seawater is then thoroughly mixed before being dispensed into the glass and plastic bottles. The filled bottles are then autoclaved at 110°C, 1.5 bar for 30 minutes. The autoclaving process removes micro-organisms, which affect the stability of the nutrient test materials. It has been demonstrated that autoclaving the test materials generates an increase in pH. We have found that after autoclaving, the pH of the nutrient test materials is within the range pH 7.5 to 8.5. The four test materials differ from each other in respect of their nutrient concentrations and the salinity of the water. The salinity of the water will be approximately 10-15 psu. One of the samples is the unspiked sample and the other samples are spiked with nutrients.

There are two bottles for each test material - one glass and one plastic. The glass bottle should only be used for the determination of TOxN, nitrite, ammonia and total-N. The plastic bottle should only be used for the determination of silicate, phosphate and total-P.

There is a separate bottle for the determination of salinity labelled Salinity ONLY.

Test materials should be stored in a refrigerator at +4°C, and analysed as soon as possible after receipt. Once the test materials are opened they should be analysed immediately.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Estuarine water (Salinity 8 - 20 psu) spiked |
| SAMPLE 2 | Estuarine water (Salinity 8 - 20 psu) spiked |
| SAMPLE 3 | Low salinity open water (Salinity 8 - 20 psu) spiked |
| SAMPLE 4 | Low salinity open water (Salinity 8 - 20 psu) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

## Determinands and concentration ranges

The following nutrients should be determined:

| Determinand | Unit | Concentration range | | Error | | AA-EQS |
|---|---|---|---|---|---|---|
| | | Estuarine water (spiked) | Low salinity open water (spiked) | Const | Prop | |
| Ammonia | µmol/L | 2—50 | 0.2—5 | 0.1 | 6.0% | |
| Nitrite | µmol/L | 0.5—25 | 0.01—2 | 0.01 | 6.0% | |
| Phosphate | µmol/L | 1—15 | 0.02—5 | 0.05 | 6.0% | |
| Salinity | psu | | | 0.001 | 0.1% | |
| Silicate | µmol/L | 5—100 | 0.5—20 | 0.1 | 6.0% | |
| Total-N | µmol/L | 10—200 | 2—20 | 0.5 | 6.0% | |
| Total-P | µmol/L | 1—20 | 0.02—2 | 0.05 | 6.0% | |
| TOxN | µmol/L | 10—100 | 0.05—15 | 0.05 | 6.0% | |

***Please report Salinity only for SAMPLE 1* in an accuracy of 3 digits**

Salinity is an indicative measurement in support of methodology.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

## Analysis

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

Only one result for the different nutrients per test material is required. The results should be expressed on the test material "as received". The concentration of the nutrients should be determined against your own calibration solutions.

## Reporting

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site.

It is not possible to report two sets of data using different methods in the same data submission forms.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional data submission form.

| ROUND | 2014 - 2 |
|---|---|
| AQ-14 DOC in seawater | |
| Test materials | SAMPLE 1, SAMPLE 2, SAMPLE 3 and SAMPLE 4 |

**Objective**

This study covers the determination of DOC in open seawater and estuarine water test materials.

**Test Materials and storage**

The test materials were prepared at the MUMM Laboratory, Ostend, Belgium, using seawater collected from the Atlantic Ocean (Seawater samples)

The seawater was filtered using a 0.45µm / 0.2µm double-membrane filter and diluted with ultrapure demineralised water for the estuarial samples. The pH of the filtered seawater was adjusted to ~pH7.2 with 0.1M hydrochloric acid and spiked to appropriate concentrations. The spiked seawater is then thoroughly mixed before being dispensed into the glass bottles. The filled bottles are then autoclaved at 110˚C, 1.5 bar for 30 minutes. The autoclaving process removes micro-organisms, which affect the stability of the DOC test materials. It has been demonstrated that autoclaving the test materials generates an increase in pH. We have found that after autoclaving, the pH of the DOC test materials is within the range pH 7.5 to 8.5. The four test materials differ from each other in respect of their DOC concentrations and the salinity of the water. The salinity of the estuarine seawater will be approximately 10-15 psu. One of the samples is the unspiked sample and the other samples are spiked with DOC.

Test materials should be stored in a refrigerator at +4˚C, and analysed as soon as possible after receipt. Once the test materials are opened they should be analysed immediately.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Seawater (Salinity > 30 psu) |
| SAMPLE 2 | Seawater (Salinity > 30 psu) spiked |
| SAMPLE 3 | Estuarine water (Salinity 8 - 20 psu) spiked |
| SAMPLE 4 | Estuarine water (Salinity 8 - 20 psu) spiked |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following nutrients should be determined:

| Determinand | Unit | Concentration range | | | Error | | AA-EQS |
|---|---|---|---|---|---|---|---|
| | | Seawater | Seawater (spiked) | Estuarine water (spiked) | Const | Prop | |
| DOC | mg C/L | 0.1—20 | 0.1—20 | 0.1—20 | 0.1 | 6.0% | |

Data assessment for unspiked samples will be carried out by calculating with a proportional error of 12.5%

**Analysis**

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

Only one result for DOC test material is required. The result should be expressed on the test material "as received". The concentration of DOC should be determined against your own calibration solutions.

**Reporting**

The result for DOC should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| AQ-3 Metals in Seawater | |
| Test materials | SAMPLE 1, SAMPLE 2, SAMPLE 3 and SAMPLE 4 |

**Objective**

This study covers the determination of metals in seawater and low salinity seawater test materials.

**Test Materials and storage**

The test materials were prepared at Alterra, Wageningen, The Netherlands. The seawater used to prepare the test materials was collected from the North Sea between Belgium and the UK, and was stored in the cold store at 7˚C in 25 litre carboys.

The test materials were prepared in bulk in a 100 litre vessel. The seawater was filtered using a 0.45 µm /0.2µm double-membrane filter. Low salinity seawater test material was prepared by diluting the seawater with ultra-pure demineralised water. All test materials are preserved with 2 ml trace metal grade nitric acid per litre of seawater. Spiked test materials were prepared by adding aqueous solutions of known trace metal concentration. Approximately 1 litre of each test material is provided. Homogeneity of the test materials is assumed, as they were prepared in bulk and thoroughly mixed, before being dispensed into 1 litre polyethylene bottles. The test materials are stable for the purposes of the exercise.

Test materials should be stored in a refrigerator at +4˚C, and analysed as soon as possible after receipt. Once the test materials are opened they should be analysed immediately.

| Code | Description |
|---|---|
| SAMPLE 1 | Seawater (Salinity > 30 psu) |
| SAMPLE 2 | Seawater (Salinity > 30 psu) spiked |
| SAMPLE 3 | Low salinity Seawater (Salinity 8 - 20 psu) spiked |
| SAMPLE 4 | Low salinity Seawater (Salinity 8 -20 psu) sample spiked with concentrations between 5 and 50 times higher than the indicative range |

**Precaution**

Some of the substances may presents a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Analysis**

Treat all test materials in the same manner as your routine samples.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

Only one result per determinand per test material is required.

**Reporting**

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site

It is not possible to report two sets of data using different methods in the same data submission forms.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange an additional data submission form.

**Determinands and concentration ranges**

The following metals should be determined:

| Determinand | Unit | Concentration range | | Error | | AA-EQS |
|---|---|---|---|---|---|---|
| | | Seawater (spiked) | Low salinity Seawater (spiked) | Const | Prop | |
| Arsenic | µg/L | 0.05—5 | 0.2—10 | 0.5 | 12.5% | |
| Boron | µg/L | 1000—5000 | 200—5000 | 0.4 | 12.5% | |
| Cadmium | µg/L | 0.001—0.5 | 0.05—1 | 0.005 | 12.5% | 0.08 |
| Chromium | µg/L | 0.01—5 | 0.5—10 | 0.1 | 12.5% | |
| Cobalt | µg/L | 0.001—0.5 | 0.01—10 | 0.2 | 12.5% | |
| Copper | µg/L | 0.05—5 | 0.2—10 | 0.2 | 12.5% | |
| Iron | µg/L | 0.05—10 | 0.2—20 | 0.4 | 12.5% | |
| Lead | µg/L | 0.0002—15 | 0.1—5 | 0.01 | 12.5% | 7.2 |
| Manganese | µg/L | 0.02—2 | 0.1—5 | 0.4 | 12.5% | |
| Nickel | µg/L | 0.2—5 | 0.1—5 | 0.2 | 12.5% | 20 |
| Silver | µg/L | 0.02—2 | 0.1—5 | 0.2 | 12.5% | |
| Tin | µg/L | 0.02—1 | 0.1—5 | 0.2 | 12.5% | |
| Vanadium | µg/L | 0.1—5 | 0.2—5 | 0.2 | 12.5% | |
| Zinc | µg/L | 0.5—20 | 0.2—10 | 0.4 | 12.5% | |

**Note that the indicative range for some determinands in the spiked low salinity sample are higher compared to the range given in the Quasimeme guide**.
**SAMPLE 4 contains concentrations of the determinands which are 5 to 50 times higher compared to the indicative range given in this protocol**.

Boron is naturally occurring at higher concentrations.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

| ROUND | 2014 - 2 |
|---|---|
| AQ-4 Mercury in Seawater | |
| Test materials | SAMPLE 1, SAMPLE 2, SAMPLE 3 and SAMPLE 4 |

**Objective**

This study covers the determination of mercury in the seawater test materials. The test materials should be analysed and one result for mercury in each test material should be reported using the data submission forms provided on the Participant site.

**Test Materials and storage**

The test materials were prepared at Alterra, Wageningen, The Netherlands. The seawater used to prepare the test materials was collected from the North Sea between Belgium and the UK, and was stored in the cold store at 7˚C in 25 litre carboys.

The test materials were prepared in bulk in a 100 litre vessel. The seawater was filtered using a 0.45 µm / 0.2 µm double-membrane filter. All test materials are preserved with 2 ml trace metal grade nitric acid per litre of seawater. Test materials were spiked with aqueous solutions of known mercury concentration.

Approximately 1 litre of each test material is provided.

Homogeneity of the test materials is assumed, as they were prepared in bulk and thoroughly mixed, before being dispensed into 1 litre glass bottles. The test materials are stable for the purposes of the exercise. Test materials should be stored in a refrigerator at +4˚C, and analysed as soon as possible after receipt. Once the test materials are opened they should be analysed immediately.

| Code | Description |
|---|---|
| SAMPLE 1 | Seawater (Salinity > 30 psu) spiked |
| SAMPLE 2 | Seawater (Salinity > 30 psu) spiked |
| SAMPLE 3 | Seawater (Salinity > 30 psu) spiked |
| SAMPLE 4 | Seawater (Salinity > 30 psu) spiked sample with concentrations between 5 and 50 times higher compared to the concentrations given in this protocol |

**Precaution**

Some of the substances may presents a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

Mercury should be determined in each test material.

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Seawater (spiked) | Const | Prop | |
| Mercury | ng/L | 0.1—100 | 0.2 | 12.5% | 50 |

**SAMPLE 4 contains concentrations of the determinands which are 5 to 50 times higher compared to the indicative range given in this protocol.**

**Analysis**

Treat all test materials in the same manner as your routine samples.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. Only one result per test material is required.

**Reporting**

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site

It is not possible to report two sets of data using different methods in the same data submission forms.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange an additional data submission form.

| ROUND | 2014 - 2 |
|---|---|
| AQ-11 | Chlorophyll-a in Seawater |
| Test materials | SAMPLE 1, SAMPLE 2 and SAMPLE 3 |

**Objective**

This study covers the determination of chlorophyll a, b, c and pheopigments in filtered seawater residue test materials.

**Test Materials and storage**

The test materials for the analysis of chlorophyll a, b, c and pheopigments were prepared at Alterra, Wageningen the Netherlands. Test materials were prepared from cultures of Isochrysis + Chaetocheros + Pyramimonas (SAMPLE 1), seawater (SAMPLE 2) and from a culture of Isochrysis only (SAMPLE 3). For each test material, the resultant damp filter paper (Whatman GF/F) was wrapped in aluminium foil, inserted into cryovial and immediately 'flash frozen' in liquid nitrogen. The test materials were stored at -80°C until the day of dispatch. The test materials were homogeneous for the purposes of the LP study.

The filter papers have been shipped on cool packs, and should be stored at -20°C, or a lower temperature, immediately upon receipt, and should be analysed as soon as possible after receipt. The cool packs will thaw slowly during transit, but this does not significantly affect the stability of the material for the purpose of this study, providing the test materials themselves are frozen immediately on receipt.

| Code | Description |
|---|---|
| SAMPLE 1 | Filtered residue from 1 litre of seawater spiked with algae culture |
| SAMPLE 2 | Filtered residue from 1 litre of estuarine seawater |
| SAMPLE 3 | Filtered residue from 1 litre of seawater |

**Precaution**

Some of the substances may presents a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following pigments should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Filtered residues | Const | Prop | |
| Chlorophyll-a | µg/L | 0.1—20 | 0.05 | 12.5% | |
| Chlorophyll-b | µg/L | 0.01—5 | 0.01 | 12.5% | |
| Chlorophyll-c | µg/L | 0.02—2.5 | 0.01 | 12.5% | |
| Pheopigments | µg/L | 0.02—2.5 | 0.01 | 12.5% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

Treat all test materials in the same manner as your routine samples. Use your normal validated methods and procedures to analyse the test materials. Only one result per determinand per test material is required. The results of each determinand should be expressed on the test materials "as

received". **Concentrations need to be calculated based on a filter prepared out of a 1 litre sample.**

Whilst you should use your normal validated methods and procedures to analyse the test materials in this study, previous QUASIMEME development exercises have shown that the best between laboratory agreement was obtained with either the Trichromatic method (Jeffrey and Humphrey 1975) or the Monochromatic method (Lorenzen 1967).

**Reporting**

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site

It is not possible to report two sets of data using different methods in the same data submission forms.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange an additional data submission form.

| ROUND | 2014 - 2 |
|---|---|
| MS-1 Trace metals in Sediment | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of metals, total organic carbon and carbonate in marine sediment test materials.

**Test Materials and storage**

Test materials were prepared by WEPAL, Wageningen, The Netherlands.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the sediment was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature prior to analysis, and analysed as soon as possible after receipt.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Sediment (open sea) |
| SAMPLE 2 | Sediment (estuarine) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following trace metals should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Sediment | Const | Prop | |
| Aluminium-AE | % | 1—10 | 0.1 | 12.5% | |
| Aluminium-RT | % | 1—10 | 0.1 | 12.5% | |
| Arsenic-AE | mg/kg | 2—50 | 1 | 12.5% | |
| Arsenic-RT | mg/kg | 2—50 | 1 | 12.5% | |
| Cadmium-AE | µg/kg | 10—2000 | 20 | 12.5% | |
| Cadmium-RT | µg/kg | 10—2000 | 20 | 12.5% | |
| Chromium-AE | mg/kg | 10—1000 | 2 | 12.5% | |
| Chromium-RT | mg/kg | 10—1000 | 2 | 12.5% | |
| Copper-AE | mg/kg | 1—500 | 1 | 12.5% | |
| Copper-RT | mg/kg | 1—500 | 1 | 12.5% | |
| Inorganic-carbonate | % | 0.05—10 | 0.05 | 12.5% | |
| Iron-AE | % | 0.5—10 | 0.1 | 12.5% | |
| Iron-RT | % | 0.5—10 | 0.1 | 12.5% | |
| Lead-AE | mg/kg | 5—500 | 2 | 12.5% | |
| Lead-RT | mg/kg | 5—500 | 2 | 12.5% | |
| Lithium-AE | mg/kg | 10—100 | 0.1 | 12.5% | |
| Lithium-RT | mg/kg | 10—100 | 0.1 | 12.5% | |
| Manganese-AE | mg/kg | 100—2000 | 0.1 | 12.5% | |
| Manganese-RT | mg/kg | 100—2000 | 0.1 | 12.5% | |
| Mercury-AE | µg/kg | 50—2500 | 10 | 12.5% | |

| | | | | | |
|---|---|---|---|---|---|
| Mercury-RT | µg/kg | 50—2500 | 10 | 12.5% | |
| Nickel-AE | mg/kg | 5—100 | 1 | 12.5% | |
| Nickel-RT | mg/kg | 5—100 | 1 | 12.5% | |
| Scandium-AE | mg/kg | 1—20 | 0.1 | 12.5% | |
| Scandium-RT | mg/kg | 1—20 | 0.1 | 12.5% | |
| TOC | % | 0.2—10 | 0.1 | 12.5% | |
| Zinc-AE | mg/kg | 20—1500 | 2.5 | 12.5% | |
| Zinc-RT | mg/kg | 20—1500 | 2.5 | 12.5% | |

RT = Real Total destructions e.g. HF-destruction, röntgen-diffraction and neutron activation
AE= Acid extractable and all other methods

*In addition to these parameters given in this table, Quasimeme will add several additional metals into the data submission form on the Participant Sites. For example, Na, Mg, P, S, K, Ca, Ti, V, Co, Ga, Rb, Sr, Mo, Sn, Cs, Ba, Ce, Ta, Tl, Th, U.* ***When Quasimeme receive sufficient reported results these additional metals will be permanently added to the programme.***

In addition, total organic carbon and inorganic carbonate should be determined for both test materials. Please note that inorganic carbonate should be reported as % carbon.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

The sediments have been dried and sieved to < 0.5 mm. Before sub-sampling, shake the bottle for at least two minutes. Repeat this re-homogenisation each time the test material is used. It is recommended that the sediments be kept in a desiccator. It is left to the discretion of the laboratory to dry the material. You are advised to check the moisture content of the sediment by drying a small portion of the sediment to constant weight at ~ 110˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

Aluminium (Al) should be determined by a total digest or non-destructive method.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material "as received".

The concentration of metals should be determined against your own calibration solutions.

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| MS-2 Chlorinated Organics in Sediment | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of chlorobiphenyls (CBs), organochlorine pesticides (OCPs) and total organic carbon in marine sediment test materials.

**Test Materials and storage**

Test materials were prepared by WEPAL, Wageningen, The Netherlands.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the sediment was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature prior to analysis, and analysed as soon as possible after receipt.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Sediment (estuarine) |
| SAMPLE 2 | Sediment (estuarine) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following Chlorinated Organics should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Sediment | Const | Prop | |
| a-HCH | µg/kg | 0.02—1 | 0.02 | 12.5% | |
| b-HCH | µg/kg | 0.05—2 | 0.025 | 12.5% | |
| CB101 | µg/kg | 0.2—50 | 0.025 | 12.5% | |
| CB105 | µg/kg | 0.1—10 | 0.025 | 12.5% | |
| CB118 | µg/kg | 0.1—50 | 0.025 | 12.5% | |
| CB138 | µg/kg | 0.2—50 | 0.025 | 12.5% | |
| CB138+CB163 | µg/kg | 0.2—50 | 0.025 | 12.5% | |
| CB153 | µg/kg | 0.2—50 | 0.025 | 12.5% | |
| CB156 | µg/kg | 0.05—5 | 0.025 | 12.5% | |
| CB180 | µg/kg | 0.1—50 | 0.025 | 12.5% | |
| CB28 | µg/kg | 0.1—50 | 0.025 | 12.5% | |
| CB31 | µg/kg | 0.1—50 | 0.025 | 12.5% | |
| CB52 | µg/kg | 0.1—50 | 0.025 | 12.5% | |
| d-HCH | µg/kg | 0.05—2 | 0.025 | 12.5% | |
| Dieldrin | µg/kg | 0.1—10 | 0.025 | 12.5% | |
| g-HCH | µg/kg | 0.05—2 | 0.025 | 12.5% | |
| HCB | µg/kg | 0.05—20 | 0.025 | 12.5% | |
| HCBD | µg/kg | 0.1—10 | 0.025 | 12.5% | |
| op'-DDT | µg/kg | 0.02—5 | 0.025 | 12.5% | |
| pp'-DDD | µg/kg | 0.1—20 | 0.025 | 12.5% | |

| | | | | | |
|---|---|---|---|---|---|
| pp'-DDE | µg/kg | 0.1—10 | 0.025 | 12.5% | |
| pp'-DDT | µg/kg | 0.1—10 | 0.025 | 12.5% | |
| TOC | % | 0.2—10 | 0.02 | 12.5% | |
| Transnonachlor | µg/kg | 0.01—2 | 0.025 | 12.5% | |

In addition total organic carbon should be determined for both test materials.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

The sediments have been dried and sieved to < 0.5 mm. Before sub-sampling, shake the bottle for at least two minutes. Repeat this re-homogenisation each time the test material is used. It is recommended that the sediments be kept in a desiccator. It is left to the discretion of the laboratory to dry the material. You are advised to check the moisture content of the sediment by drying a small portion of the sediment to constant weight at ~ 110˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material "as received".

The concentrations should be determined against your own calibration solutions.

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| MS-3 Polycyclic Aromatic Hydrocarbons in Sediment | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of PAHs and total organic carbon in marine sediment test materials.

**Test Materials and storage**

Test materials were prepared by WEPAL, Wageningen, The Netherlands.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the sediment was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature prior to analysis, and analysed as soon as possible after receipt.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Sediment (harbour) |
| SAMPLE 2 | Sediment (harbour) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following PAHs and alkylated PAHs should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Sediment | Const | Prop | |
| 1-Methylpyrene | µg/kg | 2—500 | 0.5 | 12.5% | |
| 2-Methylphenanthrene | µg/kg | 5—1000 | 0.5 | 12.5% | |
| 3,6-Dimethylphenanthrene | µg/kg | 1—500 | 0.5 | 12.5% | |
| Acenaphthene | µg/kg | 2—500 | 0.1 | 12.5% | |
| Acenaphthylene | µg/kg | 2—100 | 0.2 | 12.5% | |
| Anthracene | µg/kg | 2—500 | 0.1 | 12.5% | |
| Benzo[a]anthracene | µg/kg | 10—1500 | 0.1 | 12.5% | |
| Benzo[a]fluorene | µg/kg | 10—1000 | 0.5 | 12.5% | |
| Benzo[a]pyrene | µg/kg | 10—1500 | 0.1 | 12.5% | |
| Benzo[b]fluoranthene | µg/kg | 10—1500 | 0.5 | 12.5% | |
| Benzo[e]pyrene | µg/kg | 10—1500 | 0.2 | 12.5% | |
| Benzo[g,h,i]perylene | µg/kg | 10—1500 | 0.2 | 12.5% | |
| Benzo[k]fluoranthene | µg/kg | 10—1000 | 0.1 | 12.5% | |
| Chrysene | µg/kg | 10—1500 | 0.2 | 12.5% | |
| Chrysene+Triphenylene | µg/kg | 10—3000 | 0.2 | 12.5% | |
| Dibenz[a,h]anthracene | µg/kg | 5—500 | 0.05 | 12.5% | |
| Dibenzo[a,i]pyrene | µg/kg | | 0.5 | 12.5% | |
| Dibenzothiophene | µg/kg | 2—200 | 0.1 | 12.5% | |
| Fluoranthene | µg/kg | 20—3000 | 0.2 | 12.5% | |
| Fluorene | µg/kg | 2—300 | 0.1 | 12.5% | |

| | | | | | |
|---|---|---|---|---|---|
| Indeno[1,2,3-cd]pyrene | µg/kg | 10—1500 | 0.2 | 12.5% | |
| Naphthalene | µg/kg | 10—1500 | 0.5 | 12.5% | |
| Perylene | µg/kg | 10—500 | 0.2 | 12.5% | |
| Phenanthrene | µg/kg | 10—2000 | 0.5 | 12.5% | |
| Pyrene | µg/kg | 10—3000 | 0.2 | 12.5% | |
| TOC | % | 0.2—10 | 0.02 | 12.5% | |
| Triphenylene | µg/kg | 20—3000 | 0.5 | 12.5% | |
| C1-phenanthrenes/anthracenes | µg/kg | | 0.5 | 12.5% | |
| C2-phenanthrenes/anthracenes | µg/kg | | 0.5 | 12.5% | |
| C3-phenanthrenes/anthracenes | µg/kg | | 0.5 | 12.5% | |
| C1-pyrenes/fluoranthenes | µg/kg | | 0.5 | 12.5% | |
| C2-pyrenes/fluoranthenes | µg/kg | | 0.5 | 12.5% | |
| C1-chrysenes | µg/kg | | 0.5 | 12.5% | |
| C2-chrysenes | µg/kg | | 0.5 | 12.5% | |
| C1-benzofluoranthenes | µg/kg | | 0.5 | 12.5% | |

In addition total organic carbon should be determined for both test materials.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

The sediments have been dried and sieved to < 0.5 mm. Before sub-sampling, shake the bottle for at least two minutes. Repeat this re-homogenisation each time the test material is used. It is recommended that the sediments be kept in a desiccator. It is left to the discretion of the laboratory to dry the material. You are advised to check the moisture content of the sediment by drying a small portion of the sediment to constant weight at ~ 110˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. You may use any method with the appropriate extraction and clean-up. The final determination may be made using GC, GC-MS, HPLC etc. The method codes should be entered fully on the exercise template.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material "as received".

The concentrations should be determined against your own calibration solutions.

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 – 1 |
|---|---|
| MS-6 Organotins in Sediment | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of organotin compounds in sediment test materials.

**Test Materials and storage**

The sediment test materials were supplied by WEPAL, Wageningen.

The dry sediment test materials should be stored at room temperature, in a dry place, prior to analysis, and analysed as soon as possible after receipt.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the sediment was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Sediment (harbour) |
| SAMPLE 2 | Sediment (harbour) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following organotin compounds should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Sediment | Const | Prop | |
| Dibutyltin(DBT) | µg Sn/kg | 1—500 | 0.1 | 12.5% | |
| Diphenyltin(DPT) | µg Sn/kg | 0.1—200 | 0.1 | 12.5% | |
| Monobutyltin(MBT) | µg Sn/kg | 1—500 | 0.1 | 12.5% | |
| Monophenyltin(MPT) | µg Sn/kg | 0.1—200 | 0.1 | 12.5% | |
| Tributyltin(TBT) | µg Sn/kg | 1—500 | 0.1 | 12.5% | |
| Triphenyltin(TPT) | µg Sn/kg | 0.1—200 | 0.1 | 12.5% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

The sediments have been dried and sieved to < 0.5 mm. Before sub-sampling, shake the bottle for at least two minutes. Repeat this re-homogenisation each time the test material is used. It is recommended that the sediments be kept in a desiccator.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. Report your method codes using the Data Submission Form on your Participant site. Please check each of your method codes and update where necessary. Advise QUASIMEME of additional codes that would better describe your methodology.

Only one result per determinand per test material is required.

The results of each determinand should be expressed as Sn on the test materials “as received”. All results should be reported as µg Sn /kg weight of sediment as received

The concentration of organotins should be determined against your own calibration solutions.

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| MS-7 Brominated flame retardants in Sediment | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of brominated compounds in sediment test material.

**Test Materials and storage**

The test materials were supplied by WEPAL, Wageningen, The Netherlands.

The Sediment has been dried and sieved to < 0.5 mm. Before sub-sampling, shake the bottle for at least two minutes. Repeat this re-homogenisation each time the test material is used. It is recommended that the sediments be kept in a desiccator. It is left to the discretion of the laboratory to dry the material. You are advised to check the moisture content of the sediment by drying a small portion of the sediment to constant weight at ~ 110˚C.

All materials have been shown to be homogeneous at or below the intake mass used by the participants, and are stable for the purposes of the test.

The test materials should be stored in a dry place at room temperature in the dark, prior to analysis, and analysed as soon as possible after receipt.

| Code | Description |
|---|---|
| SAMPLE 1 | Sediment (harbour) |
| SAMPLE 2 | Sediment (open sea) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following analytes should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Sediment | Const | Prop | |
| a-HBCD | µg/kg | | 0.05 | 12.5% | |
| BDE100 | µg/kg | 0.01—10 | 0.05 | 12.5% | |
| BDE153 | µg/kg | 0.1—5 | 0.05 | 12.5% | |
| BDE154 | µg/kg | 0.01—5 | 0.05 | 12.5% | |
| BDE183 | µg/kg | 0.1—2 | 0.05 | 12.5% | |
| BDE209 | µg/kg | 20—1000 | 0.05 | 12.5% | |
| BDE28 | µg/kg | 0.01—2 | 0.05 | 12.5% | |
| BDE47 | µg/kg | 0.1—20 | 0.05 | 12.5% | |
| BDE66 | µg/kg | 0.01—10 | 0.05 | 12.5% | |
| BDE85 | µg/kg | 0.01—10 | 0.05 | 12.5% | |
| BDE99 | µg/kg | 0.1—50 | 0.05 | 12.5% | |
| b-HBCD | µg/kg | | 0.05 | 12.5% | |
| Dimethyl-TBBP-A | µg/kg | | 0.05 | 12.5% | |
| g-HBCD | µg/kg | 0.01 - 20 | 0.05 | 12.5% | |
| TBBP-A | µg/kg | | 0.05 | 12.5% | |
| Total-HBCD | µg/kg | 50—1000 | 0.05 | 12.5% | |

*Note: Indicative values for BDE209 and g-HBCD were adapted following results recently found.*

Data-assessment for biological tissue test materials will be carried out by calculating with a constant error of 0.005 µg/kg.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

The sediments have been dried and sieved to < 0.5 mm. Before sub-sampling, shake the bottle for at least two minutes. Repeat this re-homogenisation each time the test material is used. It is recommended that the sediments be kept in a desiccator.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. Report your method codes using the Data Submission Form on your Participant site. Please check each of your method codes and update where necessary. Advise QUASIMEME of additional codes that would better describe your methodology.

Only one result per determinand per test material is required.

The results of each determinand should be expressed as Sn on the test materials “as received”. All results should be reported as µg Sn /kg weight of sediment as received

The concentration of organotins should be determined against your own calibration solutions.

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| BT-1 Trace metals in Biota | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of ten trace metals, ash weight, dry weight and total lipid in biological tissue test materials.

**Test Materials and storage**

The test materials were supplied by Wageningen IMARES, Institute for Marine Resources and Ecosystem Studies, IJmuiden, The Netherlands and the Institute for Environmental Studies, Free University, Amsterdam, The Netherlands.

The jars contain approximately 50g of minced sterilised biological tissue material, to which butylhydroxytoluene (BHT) has been added as an antioxidant.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the biological tissue test materials was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature, in a dry place, prior to analysis, and analysed as soon as possible after receipt. Temperatures below 0˚C will not negatively affect the quality in the material, and we do not expect the contaminants to be affected at these temperatures. Temperatures above 30˚C may be disadvantageous for the material.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Fish fillet tissue (1 – 5% lipid content) |
| SAMPLE 2 | Mussel tissue |

**Precaution**

The jars with biological tissue test materials are filled almost to the brim, so should be opened carefully so that no tissue or moisture is spilt. The jars are filled in this way in order to eliminate as much air as possible. This minimises any degradation of the test materials in transit and storage prior to opening. The jars of biota should be cooled for a few hours at ca. 4˚C prior to opening, to eliminate possible overpressure.

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following trace metals should be determined:

| Determinand | Unit | Concentration range | | Error | | AA-EQS |
|---|---|---|---|---|---|---|
| | | Shellfish tissue | Fish muscle tissue | Const | Prop | |
| Arsenic | mg/kg | 0.2—10 | 1—10 | 0.02 | 12.5% | |
| Ash-weight | % | | | 0.1 | 12.5% | |
| Cadmium | µg/kg | 10—500 | 1—50 | 20 | 12.5% | |
| Chromium | µg/kg | 10—5000 | 50—500 | 20 | 12.5% | |
| Copper | µg/kg | 50—10000 | 100—1000 | 100 | 12.5% | |

| | | | | | | |
|---|---|---|---|---|---|---|
| Dry-weight | % | | | 0.1 | 12.5% | |
| Extractable-Lipid | % | | | 0.1 | 12.5% | |
| Lead | µg/kg | 10—1000 | 10—50 | 5 | 12.5% | |
| Mercury | µg/kg | 5—500 | 20—1000 | 20 | 12.5% | |
| Nickel | µg/kg | 10—2000 | 10—200 | 20 | 12.5% | |
| Selenium | µg/kg | 200—1000 | 200—2000 | 10 | 12.5% | |
| Silver | µg/kg | 1—500 | 0.5—50 | 5 | 12.5% | |
| Total-Lipid | % | | | 0.1 | 12.5% | |
| Zinc | mg/kg | 2—200 | 2—10 | 2 | 12.5% | |

Ash weight, dry weight and total lipid should also be determined. If you normally measure extractable lipid, there is a field in the template for reporting this measurement. This has been added following the request from a number of participants. However, we would encourage you to also report total lipid. A successful QUASH study has found no significant difference between the Smedes lipid method and the Bligh and Dyer method for total lipid determination. We therefore recommend that wherever possible you use the Smedes lipid method, as it gives better reproducibility and does not involve the use of chlorinated solvents. A copy of the Smedes lipid method protocol is included in Annex III. Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

During the sterilisation process moisture may have partly separated from the solid part of the test materials, and this may cause inhomogeneity. Therefore, prior to taking an aliquot for analysis the following treatment of the test materials is essential. After opening the test material container the complete contents should be thoroughly homogenised. As it is difficult to homogenise the sample in the tin, we recommend transferring the material to a larger container for homogenisation and sub-sampling. Aliquots should be taken immediately after homogenising, to prevent liquid lipids from separating from the solid tissue particles, causing inhomogeneity again. After sub-sampling the remaining tissue should be immediately transferred to a clean glass jar, sealed and stored at -20˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

The concentrations should be determined against your own calibration solutions.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material “as received” i.e. on a wet weight basis. If your normal method is to dry the test material prior to extraction then the dry weight must be determined and the final result calculated as wet weight using the dry weight determination. In this case the % dry weight should be reported.

% dry weight = 100 - [(wet weight - dry weight) x 100]/(wet weight)

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 – 2 |
|---|---|
| BT-2 Chlorinated Organics in Biota | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination chlorobiphenyls (CBs), organochlorine pesticides (OCPs) and total lipid in biological tissue test materials.

**Test Materials and storage**

The test materials were supplied by Wageningen IMARES, Institute for Marine Resources and Ecosystem Studies, IJmuiden, The Netherlands and the Institute for Environmental Studies, Free University, Amsterdam, The Netherlands.

The tins with biological tissue test material contain approximately 50g of minced sterilised material, to which butylhydroxytoluene (BHT) has been added as an antioxidant.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the biota was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature, in a dry place, prior to analysis, and analysed as soon as possible after receipt. Although the tins have a protective coating, moisture can cause corrosion of the surface of the tins. Apart from the moisture, temperatures below 0˚C will not negatively affect the quality in the material, and we do not expect the contaminants to be affected at these temperatures. Temperatures above 30˚C may be disadvantageous for the material.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Dab fillet tissue |
| SAMPLE 2 | Mussel tissue |

**Precaution**

The tins with biological tissue test materials are filled almost to the brim, so should be opened carefully so that no tissue or moisture is spilt. The tins are filled in this way in order to eliminate as much air as possible. This minimises any degradation of the test materials in transit and storage prior to opening. The tins of biota should be cooled for a few hours at ca. 4˚C prior to opening, to eliminate possible overpressure.

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following analytes should be determined:

| Determinand | Unit | Concentration range | | | Error | | AA-EQS |
|---|---|---|---|---|---|---|---|
| | | Fish Liver tissue and Freshwater Fish | Fish Muscle Tissue | Shellfish Tissue | Const | Prop | |
| PCB28 | µg/kg | 1—50 | 0.05—5 | 0.05—5 | 0.025 | 12.5% | |
| PCB31 | µg/kg | 1—10 | 0.03—3 | 0.03—3 | 0.025 | 12.5% | |
| PCB52 | µg/kg | 10—100 | 0.05—5 | 0.05—5 | 0.025 | 12.5% | |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| PCB101 | µg/kg | 30—300 | 0.1—20 | 0.1—20 | 0.025 | 12.5% | |
| PCB105 | µg/kg | 2—100 | 0.05—10 | 0.05—10 | 0.025 | 12.5% | |
| PCB118 | µg/kg | 20—300 | 0.2—20 | 0.2—20 | 0.025 | 12.5% | |
| PCB138+PCB163 | µg/kg | 20—600 | 0.3—30 | 0.3—30 | 0.025 | 12.5% | |
| PCB138 | µg/kg | 20—600 | 0.3—30 | 0.3—30 | 0.025 | 12.5% | |
| PCB153 | µg/kg | 50—1000 | 0.4—40 | 0.4—40 | 0.025 | 12.5% | |
| PCB156 | µg/kg | 1—40 | 0.03—10 | 0.03—10 | 0.025 | 12.5% | |
| PCB180 | µg/kg | 10—200 | 0.05—5 | 0.05—5 | 0.025 | 12.5% | |
| α-HCH | µg/kg | 0.05—5 | 0.05—5 | 0.05—5 | 0.02 | 12.5% | |
| β-HCH | µg/kg | 0.1—5 | 0.05—5 | 0.05—5 | 0.025 | 12.5% | |
| γ-HCH | µg/kg | 0.05—5 | 0.05—5 | 0.05—5 | 0.025 | 12.5% | |
| δ-HCH | µg/kg | 0.05—5 | 0.05—5 | 0.05—5 | 0.025 | 12.5% | |
| HCB | µg/kg | 2—50 | 0.02—5 | 0.02—5 | 0.025 | 12.5% | |
| HCBD | µg/kg | 0.05—5 | | | 0.025 | 12.5% | |
| Dieldrin | µg/kg | 0.5—100 | 0.2—20 | 0.2—20 | 0.025 | 12.5% | |
| pp'-DDD | µg/kg | 5—100 | 0.1—10 | 0.1—10 | 0.025 | 12.5% | |
| pp'-DDE | µg/kg | 10—500 | 0.3—30 | 0.3—30 | 0.025 | 12.5% | |
| op'-DDT | µg/kg | 0.1—2 | 0.01—1 | 0.01—1 | 0.025 | 12.5% | |
| pp'-DDT | µg/kg | 0.1—10 | 0.1—10 | 0.1—10 | 0.025 | 12.5% | |
| Transnonachlor | µg/kg | 0.2—40 | 0.02—10 | 0.02—10 | 0.025 | 12.5% | |
| Total-Lipid | % | | | | 0.1 | 12.5% | |
| Extractable-Lipid | % | | | | 0.1 | 12.5% | |

In addition total lipid should be determined. If you normally measure extractable lipid, there is a field in the template for reporting this measurement. This has been added following the request from a number of participants. However, we would encourage you to also report total lipid. A successful QUASH study has found no significant difference between the Smedes lipid method and the Bligh and Dyer method for total lipid determination. We therefore recommend that wherever possible you use the Smedes lipid method, as it gives better reproducibility and does not involve the use of chlorinated solvents. A copy of the Smedes lipid method protocol is included in Annex III.
Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

During the sterilisation process moisture may have partly separated from the solid part of the test materials, and this may cause inhomogeneity. Therefore, prior to taking an aliquot for analysis the following treatment of the test materials is essential. After opening the test material container the complete contents should be thoroughly homogenised. As it is difficult to homogenise the sample in the tin, we recommend transferring the material to a larger container for homogenisation and sub-sampling. Aliquots should be taken immediately after homogenising, to prevent liquid lipids from separating from the solid tissue particles, causing inhomogeneity again. After sub-sampling the remaining tissue should be immediately transferred to a clean glass jar, sealed and stored at -20˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

The concentrations should be determined against your own calibration solutions.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material "as received" i.e. on a wet weight basis. If your normal method is to dry the test material prior to extraction then the dry weight must be determined and the final result calculated as wet weight using the dry weight determination. In this case the % dry weight should be reported.

% dry weight = 100 - [(wet weight - dry weight) x 100]/(wet weight)

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| BT-4 Polycyclic Aromatic Hydrocarbons in Biota | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of PAHs and total lipid in biological tissue test materials.

**Test Materials and storage**

The test materials were supplied by Wageningen IMARES, Institute for Marine Resources and Ecosystem Studies, IJmuiden, The Netherlands and the Institute for Environmental Studies, Free University, Amsterdam, The Netherlands.

The tins contain approximately 50g of minced sterilised biological tissue material, to which butylhydroxytoluene (BHT) has been added as an antioxidant. Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the biota was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature, in a dry place, prior to analysis, and analysed as soon as possible after receipt. Although the tins have a protective coating, moisture can cause corrosion of the surface of the tins. Apart from the moisture, temperatures below 0˚C will not negatively affect the quality in the material, and we do not expect the contaminants to be affected at these temperatures. Temperatures above 30˚C may be disadvantageous for the material.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Mussel tissue |
| SAMPLE 2 | Mussel tissue |

**Precaution**

The tins with biological tissue are filled almost to the brim, so should be opened carefully so that no tissue or moisture is spilt. The tins are filled in this way in order to eliminate as much air as possible. This minimizes any degradation of the test materials in transit and storage prior to opening. The tins of biota should be cooled for a few hours at ca. 4˚C prior to opening, to eliminate possible overpressure. Opening the bottom of the tin, instead of the top, may also help to prevent spillage of moisture.

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following PAHs should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Shellfish tissue | Const | Prop | |
| 1-Methylpyrene | µg/kg | | 2 | 12.5% | |
| 2-Methylphenanthrene | µg/kg | 0.2—5 | 2 | 12.5% | |
| 3,6-Dimethylphenanthrene | µg/kg | 0.2—2 | 0.5 | 12.5% | |
| Acenaphthene | µg/kg | 0.5—100 | 0.2 | 12.5% | |
| Acenaphthylene | µg/kg | 0.2—5 | 0.2 | 12.5% | |

| | | | | | |
|---|---|---|---|---|---|
| Anthracene | µg/kg | 0.2—10 | 0.2 | 12.5% | |
| Benzo[a]anthracene | µg/kg | 0.2—10 | 0.2 | 12.5% | |
| Benzo[a]fluorene | µg/kg | | 0.5 | 12.5% | |
| Benzo[a]pyrene | µg/kg | 0.2—5 | 0.2 | 12.5% | |
| Benzo[b]fluoranthene | µg/kg | 0.2—10 | 0.2 | 12.5% | |
| Benzo[e]pyrene | µg/kg | 0.2—10 | 0.2 | 12.5% | |
| Benzo[g,h,i]perylene | µg/kg | 0.2—5 | 0.2 | 12.5% | |
| Benzo[k]fluoranthene | µg/kg | 0.2—5 | 0.2 | 12.5% | |
| Chrysene | µg/kg | 0.2—20 | 0.2 | 12.5% | |
| Chrysene+Triphenylene | µg/kg | 0.2—20 | 0.2 | 12.5% | |
| Dibenz[a,h]anthracene | µg/kg | 0.2—2 | 0.1 | 12.5% | |
| Dibenzo[a,i]pyrene | µg/kg | | 0.5 | 12.5% | |
| Dibenzothiophene | µg/kg | 0.2—5 | 0.5 | 12.5% | |
| Extractable-Lipid | % | | 0.1 | 12.5% | |
| Fluoranthene | µg/kg | 5—50 | 0.2 | 12.5% | |
| Fluorene | µg/kg | 1—50 | 0.2 | 12.5% | |
| Indeno[1,2,3-cd]pyrene | µg/kg | 0.2—5 | 0.2 | 12.5% | |
| Naphthalene | µg/kg | 1—100 | 0.2 | 12.5% | |
| Perylene | µg/kg | 0.1—5 | 0.5 | 12.5% | |
| Phenanthrene | µg/kg | 2—50 | 0.2 | 12.5% | |
| Pyrene | µg/kg | 1—20 | 0.2 | 12.5% | |
| Total-Lipid | % | | 0.1 | 12.5% | |
| Triphenylene | µg/kg | | 5 | 12.5% | |

In addition total lipid should be determined. If you normally measure extractable lipid, there is a field in the template for reporting this measurement. This has been added following the request from a number of participants. However, we would encourage you to also report total lipid. A successful QUASH study has found no significant difference between the Smedes lipid method and the Bligh and Dyer method for total lipid determination. We therefore recommend that wherever possible you use the Smedes lipid method, as it gives better reproducibility and does not involve the use of chlorinated solvents. A copy of the Smedes lipid method protocol is included in Annex III.

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

During the sterilisation process moisture may have partly separated from the solid part of the test materials, and this may cause inhomogeneity. Therefore, prior to taking an aliquot for analysis the following treatment of the test materials is essential. After opening the test material container the complete contents should be thoroughly homogenised. As it is difficult to homogenise the sample in the tin, we recommend transferring the material to a larger container for homogenisation and sub-sampling. Aliquots should be taken immediately after homogenising, to prevent liquid lipids from separating from the solid tissue particles, causing inhomogeneity again. After sub-sampling the remaining tissue should be immediately transferred to a clean glass jar, sealed and stored at -20˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

The concentrations should be determined against your own calibration solutions.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material “as received” i.e. on a wet weight basis. If your normal method is to dry the test material prior to extraction then the dry weight must be determined and the final result calculated as wet weight using the dry weight determination. In this case the % dry weight should be reported.

% dry weight = 100 - [(wet weight - dry weight) X 100]/(wet weight)

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| BT-8 Organotins in Biota | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of organotin compounds in biological tissue test materials.

**Test Materials and storage**

The test materials were supplied by Wageningen IMARES, Institute for Marine Resources and Ecosystem Studies, IJmuiden, The Netherlands and the Institute for Environmental Studies, Free University, Amsterdam, The Netherlands.

The tins contain approximately 50g of minced sterilised biological tissue material, to which butylhydroxytoluene (BHT) has been added as an antioxidant. Each batch of material was prepared in bulk. The level of within and between sample homogeneity for the biota was determined. All materials have been shown to be homogeneous at or below the intake mass used by the participants, and stable for the purposes of the test.

The test materials should be stored at room temperature, in a dry place, prior to analysis, and analysed as soon as possible after receipt. Although the tins have a protective coating, moisture can cause corrosion of the surface of the tins. Apart from the moisture, temperatures below 0°C will not negatively affect the quality in the material, and we do not expect the contaminants to be affected at these temperatures. Temperatures above 30°C may be disadvantageous for the material.

Treat all test materials in the same manner as your routine samples.

| Code | Description |
|---|---|
| SAMPLE 1 | Mussel tissue |
| SAMPLE 2 | Mussel tissue |

**Precaution**

The tins or jars with biological tissue are filled almost to the brim, so should be opened carefully so that no tissue or moisture is spilt. The tins or jars are filled in this way in order to eliminate as much air as possible. This minimizes any degradation of the test materials in transit and storage prior to opening. The tins or jars of biota should be cooled for a few hours at ca. 4°C prior to opening, to eliminate possible overpressure.

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The following analytes should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Biota | Const | Prop | |
| Dibutyltin(DBT) | µg Sn/kg | 1—100 | 0.1 | 12.5% | |
| Diphenyltin(DPT) | µg Sn/kg | | 0.1 | 12.5% | |
| Monobutyltin(MBT) | µg Sn/kg | 5—30 | 0.1 | 12.5% | |
| Monophenyltin(MPT) | µg Sn/kg | | 0.1 | 12.5% | |
| Tributyltin(TBT) | µg Sn/kg | 2—50 | 0.1 | 12.5% | |
| Triphenyltin(TPT) | µg Sn/kg | | 0.1 | 12.5% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

During the sterilisation process moisture may have partly separated from the solid part of the test materials, and this may cause inhomogeneity. Therefore, prior to taking an aliquot for analysis the following treatment of the test materials is essential. After opening the test material container the complete contents should be thoroughly homogenised. As it is difficult to homogenise the sample in the tin, we recommend transferring the material to a larger container for homogenisation and sub-sampling. Aliquots should be taken immediately after homogenising, to prevent liquid lipids from separating from the solid tissue particles, causing inhomogeneity again. After sub-sampling the remaining tissue should be immediately transferred to a clean glass jar, sealed and stored at -20˚C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

The concentrations should be determined against your own calibration solutions.

Only one result per determinand per test material is required. The results of each determinand should be expressed as Sn on the test material "as received" i.e. on a wet weight basis (µg Sn/kg).

If your normal method is to dry the test material prior to extraction then the dry weight must be determined and the final result calculated as wet weight using the dry weight determination. In this case the % dry weight should be reported.

% dry weight = 100 - [(wet weight - dry weight) x 100]/(wet weight)

**Reporting**

One result for each determinand in each test material should be reported using the Data Submission Form which is placed on your Participant site. It is not possible to report two sets of data using different methods on the same exercise Data Submission Form.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise Data Submission Form.

| ROUND | 2014 - 2 |
|---|---|
| BT-9 Brominated Flame Retardants in Biota | |
| Test materials | SAMPLE 1 and SAMPLE 2 |

**Objective**

This study covers the determination of brominated compounds in biota.

**Test Materials and storage**

The test materials were supplied by Wageningen IMARES, Institute for Marine Resources and Ecosystem Studies, IJmuiden, The Netherlands and the Institute for Environmental Studies, Free University, Amsterdam, The Netherlands.

The tins or jars contains approximately 50g of minced sterilised material, to which butylhydroxytoluene (BHT) has been added as an antioxidant. Representative sub samples were dispensed into tins and sterilised by autoclaving. Each batch of material was prepared in bulk.

Homogeneity, Stability and Storage

All materials have been shown to be homogeneous at or below the intake mass used by the participants, and are stable for the purposes of the test.

The test materials should be stored at room temperature, prior to analysis, and analysed as soon as possible after receipt.

| Code | Description |
|---|---|
| SAMPLE 1 | Mussel tissue |
| SAMPLE 2 | Fish fillet tissue 1 – 5% fat (freshwater fish) |

**Precaution**

The tins or jars with biological tissue are filled almost to the brim, so should be opened carefully so that no tissue or moisture is spilt. The tins or jars are filled in this way in order to eliminate as much air as possible. This minimises any degradation of the test materials in transit and storage prior to opening. The tins or jars of biota should be cooled for a few hours at ca. 4°C prior to opening, to eliminate possible overpressure.

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

## Determinands and concentration ranges

The following analytes should be determined:

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Biota | Const | Prop | |
| BDE28 | µg/kg | 0.001—1 | 0.05 | 12.5% | |
| BDE47 | µg/kg | 0.05—20 | 0.05 | 12.5% | |
| BDE66 | µg/kg | 0.01—10 | 0.05 | 12.5% | |
| BDE85 | µg/kg | 0.01—10 | 0.05 | 12.5% | |
| BDE99 | µg/kg | 0.01—10 | 0.05 | 12.5% | |
| BDE100 | µg/kg | 0.005—2 | 0.05 | 12.5% | |
| BDE153 | µg/kg | 0.01—1 | 0.05 | 12.5% | |
| BDE154 | µg/kg | 0.001—1 | 0.05 | 12.5% | |
| BDE183 | µg/kg | 0.001—0.1 | 0.05 | 12.5% | |
| BDE209 | µg/kg | 0.01—0.1 | 0.05 | 12.5% | |
| TBBP-A | µg/kg | | 0.05 | 12.5% | |
| Dimethyl-TBBP-A | µg/kg | | 0.05 | 12.5% | |
| α-HBCD | µg/kg | | 0.05 | 12.5% | |
| β-HBCD | µg/kg | | 0.05 | 12.5% | |
| δ-HBCD | µg/kg | | 0.05 | 12.5% | |
| Total-HBCD | µg/kg | | 0.05 | 12.5% | |

Data-assessment for biological tissue test materials will be carried out by calculating with a constant error of 0.005 µg/kg.

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

## Analysis

During the sterilisation process of the biological tissue test material moisture may have partly separated from the solid part of the test materials, and this may cause inhomogeneity. Therefore, prior to taking an aliquot for analysis the following treatment of the test materials is essential. After opening the test material container the complete contents should be thoroughly homogenised. As it is difficult to homogenise the sample in the jar, we recommend transferring the material to a larger container for homogenisation and sub-sampling. Aliquots should be taken immediately after homogenising, to prevent liquid lipids from separating from the solid tissue particles, causing inhomogeneity again. After sub-sampling the remaining tissue should be immediately transferred to a clean glass jar, sealed and stored at -20°C.

Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. Recovery values of over 100% should not be used to correct the data.

Only one result per determinand per test material is required. The results of each determinand should be expressed on the test material "as received" i.e. on a wet weight basis. If your normal method is to dry the test material prior to extraction then the dry weight must be determined and the final result calculated as wet weight using the dry weight determination. In this case the % dry weight should be reported.

% dry weight = 100 - [(wet weight - dry weight) $*$ 100]/(wet weight)

You may wish to use two different GC columns of different polarity for the determination of the BDEs. Use your own judgement to report the best result. The column used for this result should be reported.

## Reporting

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site.

It is not possible to report two sets of data using different methods in the same data submission forms.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange to send you an additional data submission form.

| ROUND | 2014 - 2 |
|---|---|
| BT-7 ASP Shellfish Toxins | |
| Test materials | SAMPLE 1, SAMPLE 2 and SAMPLE 3 |

**Objective**

This study covers the determination of amnesic shellfish toxins domoic acid and epidomoic acid (as a racemic mixture) in standard solution and shellfish tissue test materials.

**Test Materials and storage**

The test materials were supplied by the Marine Institute, Galway, Republic of Ireland.

- SAMPLE 1 is a ASP standard solution.

- SAMPLE 2 is an oyster tissue homogenate supplied in a plastic vial.

- SAMPLE 3 is a scallop tissue homogenate supplied in a plastic vial.

For SAMPLE 2 and SAMPLE 3, each vial contains sufficient material for one-shot analysis of domoic and epidomoic acid.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity and stability was determined. All materials have been shown to be homogeneous at or below the intake mass normally used, and stable for the purposes of the test.

Begin the analysis as soon as possible, preferably within 7 days of receipt.

The shellfish tissue homogenates (contained in 5ml plastic vials) should be stored at -20˚C, or a lower temperature, immediately upon receipt, until analysis.

The standard solution SAMPLE 1 should be stored in the refrigerator at ca 4˚C immediately upon receipt, until analysis

The test materials have been shipped on cool packs. The cool packs will thaw slowly during transit, but this does not significantly affect the stability of the material for the purpose of this study, providing the test materials themselves are stored as directed, immediately on receipt.

| Code | Description |
|---|---|
| SAMPLE 1 | Standard Solution |
| SAMPLE 2 | Shellfish tissue (Oyster) |
| SAMPLE 2 | Shellfish tissue (Scallop) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

Report the sum of the domoic acid and epidomoic acid as a racemic mixture.

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | Shellfish tissue | Const | Prop | |
| Domoic+Epidomoic | mg/kg | | 0.1 | 12.5% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Analysis**

It is advisable to analyse the test materials as soon as possible after receipt. Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. One result per test material is required, for the sum of domoic and epidomoic acid as a racemic mixture. The results of each determinand should be expressed on the test material "as received" i.e. on a wet weight basis. The concentrations should be determined against your own calibration solutions.

Each vial contains sufficient quantity of homogenate for one analysis. The whole transferable contents of each vial should be extracted, and one result reported for the sum of the two isomers. To transfer the contents into a preweighed or tared extraction tube, the vial should be fully defrosted, vortex-mixed and the contents poured into the desired container.

All results should be reported in mg/kg on the basis of wet weight of the test material as provided. The density of the standard solution is 0.9853 g/ml at 22°C. The weight of shellfish tissue test materials should be determined prior to analysis.

**Reporting**

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site

It is not possible to report two sets of data using different methods in the same data submission forms.

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange an additional data submission form.

| ROUND | 2014 - 2 |
|---|---|
| BT-11 Lipophilic Shellfish Toxins | |
| Test materials | SAMPLE 1, SAMPLE 2, SAMPLE3, SAMPLE 4 and SAMPLE 5 |

**Objective**

This study covers the determination of lipophilic toxins in shellfish tissue.

**Test Materials and storage**

The test materials were supplied by the Marine Institute, Galway, Republic of Ireland.

Each vial contains sufficient material for one-shot analysis of OA, AZA, YTX and PTX-group toxins..

Each batch of material was prepared in bulk. The level of within and between sample homogeneity and stability was determined. All materials have been shown to be homogeneous at the intake mass normally used, and stable for the purposes of the test.

Begin the analysis as soon as possible, preferably within 7 days of receipt.

All materials (contained either in ampoules or in 5ml plastic vials) should be stored at -20°C, or a lower temperature, immediately upon receipt, until analysis.

The test materials have been shipped on cool packs. The cool packs will thaw slowly during transit, but this does not significantly affect the stability of the material for the purpose of this study, providing the test materials themselves are stored as directed, immediately on receipt.

| Code | Description |
|---|---|
| SAMPLE 1 | AZA Standard Solution |
| SAMPLE 2 | Lipophilic Standard Solution |
| SAMPLE 3 | DSP/AZP Extract |
| SAMPLE 4 | Shellfish tissue (mussel) |
| SAMPLE 5 | Shellfish tissue (mussel) |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

Determinands

a) Methods based on chromatographic separation techniques (e.g. LC-FD, or LC-MS):

Please report lipophilic toxins (if detected) as follows:

- Free OA-group toxins, OA, DTX-1, DTX-2 (pre-hydrolysis) individually and their sum (standard solutions, extracts and tissues).
- Total OA-group toxins, OA, DTX-1, DTX-2 (post-hydrolysis) individually and their sum (extracts and tissues only).
- PTX-group toxins, PTX-1 and PTX-2 individually (standard solutions, extracts and tissues).
- Total OA-group and PTX-group toxins, sum of OA, DTX-1, DTX-2 (post-hydrolysis), PTX-1 and PTX-2 (extracts and tissues only).
- AZA-group toxins, AZA-1, AZA-2 and AZA-3 individually and their sum (standard solutions, extracts and tissues).

- YTX-group toxins, YTX, homo-YTX, 45-OH-YTX and 45-OH-homo-YTX individually and their sum (standard solutions, extracts and tissues).

Please note, for the OA-group toxins, there is no result reported for the ester-forms themselves, only for free toxins and the sum of free toxins plus esters.

b) Methods based on determination of the sum of OA-equivalents present (e.g. PP2a):

For the standard solution, report the sum of OA-equivalents as free toxins (without hydrolysis), and the sum of OA-equivalents post hydrolysis. This means there is no result reported for the esters-forms themselves, only for free toxins and the sum of free toxins plus esters.

If you do not analyse for one of the determinands, eg. DTX-1 or DTX-2, please do not report the sum of OA+DTX-1+DTX-2. Equally if you do not carry out hydrolysis or determination of DTX-1 or DTX-2 post-hydrolysis, please do not report the sum of hydrolysed results.

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | | Const | Prop | |
| AZA-1 | µg/kg | | 0.1 | 12.5% | |
| AZA-2 | µg/kg | | 0.1 | 12.5% | |
| AZA-3 | µg/kg | | 0.1 | 12.5% | |
| AZA-total | µg AZA-eq./kg | | 0.1 | 12.5% | |
| Free-DTX1 | µg/kg | | 0.1 | 12.5% | |
| Free-DTX2 | µg/kg | | 0.1 | 12.5% | |
| Free-Okadaic-Acid | µg/kg | | 0.1 | 12.5% | |
| Total-free-OA+DTX-1+DTX-2 | µg OA-eq./kg | | 0.1 | 12.5% | |
| Total-DTX1 | µg/kg | | 0.1 | 12.5% | |
| Total-DTX2 | µg/kg | | 0.1 | 12.5% | |
| Total-Okadaic Acid | µg/kg | | 0.1 | 12.5% | |
| Total-hy-OA+DTX1+DTX2 | µg OA-eq./kg | | 0.1 | 12.5% | |
| PTX-1 | µg/kg | | 0.1 | 12.5% | |
| PTX-2 | µg/kg | | 0.1 | 12.5% | |
| Total-OA-group and PTX-group | µg OA-eq./kg | | 0.1 | 12.5% | |
| YTX | mg/kg | | 0.1 | 12.5% | |
| homo-YTX | mg/kg | | 0.1 | 12.5% | |
| 45-OH-YTX | mg/kg | | 0.1 | 12.5% | |
| 45-OH-homo-YTX | mg/kg | | 0.1 | 12.5% | |
| Total-YTX-group | mg YTX-eq./kg | | 0.1 | 12.5% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

**Toxicity Equivalency Factors**

Total toxicity equivalence for each of the biotoxin groups should be calculated using toxicity equivalency factors (TEFs) as recommended by EFSA, which are contained in the following table:

| Toxin Group | Analogue | TEF |
|---|---|---|
| OA-group | OA | 1 |
| | DTX-1 | 1 |
| | DTX-2 | 0.6 |
| AZA-group | AZA-1 | 1 |
| | AZA-2 | 1.8 |
| | AZA-3 | 1.4 |

| | | |
|---|---|---|
| PTX-group | PTX-1 | 1 |
| | PTX-2 | 1 |
| YTX-group | YTX | 1 |
| | homo-YTX | 1 |
| | 45-OH-YTX | 1 |
| | 45-OH-homo-YTX | 0.5 |

**Analysis**

It is advisable to analyse the test materials as soon as possible after receipt. Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery.

One determination of each test material is required, for each determinand. The results of each determinand should be expressed on the test material "as received" i.e. on a wet weight basis. The concentrations should be determined against your own calibration solutions.

If you routinely conduct analyses by more than one technique you may report multiple sets of data.

You should inform the QUASIMEME office staff, who will arrange to send you an additional exercise template file. It is not possible to report two sets of data using different methods on the same exercise template.

The standard solution ampoules contain at least 500 µl, sufficient for 1 injection of a solution into a LC or 1 analyses of the solution by an assay.

Each vial contains sufficient quantity of extract or homogenate for one analysis. The whole transferable contents of each vial should be extracted. To transfer the contents into a preweighed or tared extraction tube, the vial should be fully defrosted, vortex-mixed and the contents poured into the desired container.

Please note all test materials should be stored in the freezer at ca -20°C or less between analyses.

All results should be reported in µg/kg (YTX-group toxins in mg/kg) on the basis of wet weight of the test material as provided. The density of the standard solutions are 0.7918 g/ml (MeOH) and the density of the lipophilic extract is 0.834g/ml. For the tissues, the weight of material should be determined prior to analysis.

Please note that if your laboratory does not report on a given analogue, e.g. DTX-1, then your laboratory should not report the sum of toxins, since this will give 2 z-scores out of line and will possibly make data-analysis more difficult for the remaining laboratories which did determine this analyte.

**Reporting**

One result for each determinand in each test material should be reported using the data submission forms which are placed at the Participant site

| ROUND | 2014 - 2 |
|---|---|
| BT-12 PSP Shellfish Toxins | |
| Test materials | SAMPLE 1, SAMPLE 2, SAMPLE 3 and SAMPLE 4 |

**Objective**

This study covers the determination of paralytic shellfish toxins in shellfish tissue.

**Test Materials and storage**

The test materials were supplied by the Marine Institute, Galway, Republic of Ireland.

Shellfish tissue test materials are supplied in a plastic 5ml vial, each vial contains sufficient material for one-shot analysis of the paralytic shellfish toxins.

Each batch of material was prepared in bulk. The level of within and between sample homogeneity and stability was determined. All materials have been shown to be homogeneous at or below the intake mass normally used, and stable for the purposes of the test.

Begin the analysis as soon as possible, preferably within 7 days of receipt.

The shellfish tissue test materials should be stored at -20˚C, or a lower temperature, immediately upon receipt, until analysis. The test materials have been shipped on cool packs. The cool packs will thaw slowly during transit, but this does not significantly affect the stability of the material for the purpose of this study, providing the test materials themselves are stored as directed, immediately on receipt.

| Code | Description |
|---|---|
| SAMPLE 1 | Mussel tissue |
| SAMPLE 2 | Oyster tissue |
| SAMPLE 3 | Mussel tissue |
| SAMPLE 4 | Oyster tissue |

**Precaution**

Some of the substances may present a health hazard and can be biologically active. Please ensure that your analytical and handling procedures for this material have been fully assessed for safety and that all specified precautions are taken.

**Determinands and concentration ranges**

The final (total toxicity) result for each test material should be reported as µg STX dihydrochloride equivalents/kg (such that HPLC, MBA, LC-MS/MS and ELISA results are comparable).
Participants using HPLC methods should also report each PSP analogue identified and give individual analogue concentrations in µmol/kg sample.

Participants using HPLC methods should use the specific toxicities as they appear in the EFSA Scientific Opinion of the Panel on Contaminants in the Food Chain for Marine Biotoxins in shellfish: STX group (see below).

| Determinand | Unit | Concentration range | Error | | AA-EQS |
|---|---|---|---|---|---|
| | | | Const | Prop | |
| 11-OH-STX | µmol/kg | | 0.1 | 12.5% | |
| C1 | µmol/kg | | 0.1 | 12.5% | |
| C2 | µmol/kg | | 0.1 | 12.5% | |
| C3 | µmol/kg | | 0.1 | 12.5% | |
| C4 | µmol/kg | | 0.1 | 12.5% | |
| dc-GTX1 | µmol/kg | | 0.1 | 12.5% | |
| dc-GTX2 | µmol/kg | | 0.1 | 12.5% | |
| dc-GTX3 | µmol/kg | | 0.1 | 12.5% | |
| dc-GTX4 | µmol/kg | | 0.1 | 12.5% | |
| dc-NEO | µmol/kg | | 0.1 | 12.5% | |
| dc-STX | µmol/kg | | 0.1 | 12.5% | |
| GTX1 | µmol/kg | | 0.1 | 12.5% | |
| GTX2 | µmol/kg | | 0.1 | 12.5% | |
| GTX3 | µmol/kg | | 0.1 | 12.5% | |
| GTX4 | µmol/kg | | 0.1 | 12.5% | |
| GTX5 | µmol/kg | | 0.1 | 12.5% | |
| GTX-6 | µmol/kg | | 0.1 | 12.5% | |
| NEO | µmol/kg | | 0.1 | 12.5% | |
| STX | µmol/kg | | 0.1 | 12.5% | |
| Total toxicity | µgSTXdiHCl-eq/kg | | 2 | 12.5% | |

Results should be reported for as many of these determinands as possible. Take this opportunity either to develop your methodology or check your performance on the less common determinands.

*TEFs recommended by the EFSA*

| Determinand | TEF |
|---|---|
| STX | 1 |
| NeoSTX | 1 |
| GTX-1 | 1 |
| GTX-2 | 0.4 |
| GTX-3 | 0.6 |
| GTX-4 | 0.7 |
| GTX-5 | 0.1 |
| GTX-6 | 0.1 |
| C2 | 0.1 |
| C4 | 0.1 |
| dc-STX | 1 |
| dc-NeoSTX | 0.4 |
| dc-GTX-2 | 0.2 |
| dc-GTX-3 | 0.4 |

## Analysis

It is advisable to analyse the test materials as soon as possible after receipt. Use your normal validated methods and procedures to analyse the test materials. This may include correcting for blanks and for recovery. One result per test material is required, for the STX analogues individually and as total STX-equivalents. The results of each determinand should be expressed on the test material "as received" i.e. on a wet weight basis. The concentrations should be determined against your own calibration solutions.

Each vial contains sufficient quantity of homogenate for one analysis. The whole transferable contents of each vial should be extracted, and one result reported for the sum of the two isomers (If participants are using the AOAC 2005.06 method then those toxins that co-elute (eg GTX1 and GTX-4, GTX-2 and GTX-3, dcGTX-2 and dcGTX-3 or C-1 and C-2) must be reported using the higher toxicity factor of the two isomers. To transfer the contents into a preweighed or tared extraction tube, the vial

should be fully defrosted, vortex-mixed and the contents poured into the desired container. The weight of the shellfish tissue test material should be determined prior to analysis.

**Reporting**

One result for each determinand in each test material should be reported using the data submission forms which are placed on the Participant site

It is not possible to report two sets of data using different methods in the same data submission forms.

## Reporting of Results and Analytical Methods

If you routinely conduct analyses by more than one technique you may report multiple sets of data. You should inform the QUASIMEME office staff, who will arrange an additional data submission form.

### Units

The units of measurement are given in the data submission forms. Ensure that the concentration of each determinand is reported in the units given. This may differ from your normal units for reporting; it is essential that all data reported are comparable. It is not possible for you to alter the units for reporting in the data submission forms.

The precision of the reported results should reflect the level of uncertainty of the measurement in your laboratory

### Reporting Left Censored Values

If the concentration of a determinand is below the detection limit of your method, you may wish to report the value as less than the detection limit. To do this, you should report your detection limit, either as a negative number or preceded by the "less-than" symbol, <. I.e. to report a value less than a detection limit of 10, report either "-10" or "<10". The system will identify either of these formats as left censored ("less-than") values. Left censored values are included in the statistical evaluation of the data, and in the reports.

### Method Codes

Method codes are supplied as part of the data submission forms. Report all of the requested method codes. If the method codes in any section do not adequately describe your analytical method, select "Other" from the method code list, and provide additional information on your method, electronically, when you return your data.

### Return of Data

Upload all analytical data to the QUASIMEME site only with the data submission forms. This allows a rapid and accurate transfer of your data and an early report to you. Additional information and comments may be provided as attached files.

Only data submitted using the data submission forms can be included in the assessment. Return the results to the QUASIMEME Project Office in Wageningen no later than 1 January 2015. Data arriving after this deadline may not be entered into the database or appear in the report.

If you have further information on additional methods used or specific ways in which we can improve the data transfer, please inform the QUASIMEME Project Office. Your co-operation is appreciated and will help the assessors in the data analysis and in providing appropriate advice in case of any analytical difficulties.

Please observe the following guidelines, to reduce the need for additional checks, replies and enquires:

Data should only be submitted to the QUASIMEME Project Office when all quality checks have been made. If data are submitted beyond the deadline, they might not be included in the report. Data submitted after the issue of the report will not be included in the report, and these data will also not be included as part of the consensus value. Any certificate prepared with data submitted late will include the statement "Data submitted after report issued". No data will be re-entered into the database after the report is issued. No data will be changed in the database UNLESS there is evidence that QUASIMEME or data transfer has caused an error. In such cases QUASIMEME will undertake a quality query to investigate the problem and inform the participant of the outcome of the Query.

The assigned values will be calculated based on the assessment of all data returned, using the Cofino model. The report for each study, including each laboratory's individual assessment and z-scores, will

be distributed to participants no later than 1 February 2015. Background information on the data assessment will be provided with the reports.

**Collusion and Falsification of Results**

QUASIMEME accepts that most participants operate with professional integrity and that data returned as part of the LP studies are correct and are submitted without interference or collusion. However, in some circumstances, data or information may be influenced by, for example, (i) repeated analyses and submitting mean data, or (ii) collaboration with colleagues undertaking the same study.

QUASIMEME checks for evidence of collusion and confirm to all participants that such activity is contrary to professional scientific conduct and will only nullify the benefits of the LP studies to accreditation bodies and analysts alike.

QUASIMEME reserves the right to withdraw participation of any institute who, in the opinion of the Scientific Assessment Board, has submitted data following collusion or falsification. This statement is made as a formal requirement for accreditation for Laboratory Performance Studies under G13: 2000 3.9.

# ANNEX 1 Notification of damaged test materials.

**You do not need to notify QUASIMEME if the test materials arrived in good condition**

Laboratory Code : .....

Damaged container number : .....

Loss of weight container number : .....

I request a new test material for :..... Because : .....

Date : .....

Signature :.....

Name of participant :.....

Name and address of institute : .....

.....

.....

Telephone number :.....

Fax number :.....

Return this form to :

**QUASIMEME Project Office**
**Wageningen UR**
**P.O. Box 8005**
**NL-6700 EC Wageningen**
**The Netherlands**

**Fax No : +31(0)317 486 546**
**E-mail : QUASIMEME@wur.nl**

# ANNEX 2 Instructions for login into new Participant's Site

Login to http://www.participants.wepal.nl/

Ask the Quasimeme project office when the login information is unknown

Select the correct year

Select the correct round

Select the correct exercise

Enter your results and method information into the data submission form

Lower than results will be automatically transferred into – values.

Data results turn green when saved and red if a mistake is made.

There is a .pdf option if you wish to print your results for your records

# ANNEX 3 Total Lipid Extraction According to Smedes

This method is based on research carried out by Foppe Smedes.

See : Determination of total lipid using non-chlorinated solvents

Smedes, F., Analyst 124 (1999) : 1711-1718.

**Instruments and Chemicals**

- Balance with a precision of 0.1 mg
- Ultra Turrax
- Centrifuge capable of holding 100 ml tubes or glass jar at a speed of 2000 rpm[1]
- Heated waterbath with condensers.
- Evaporation flasks in suitable shape and size
- Pipettes
- Deionised water
- Isopropanol
- Cyclohexane
- Solution of 13 % (w/w) isopropanol in cyclohexane.

**Procedure**

- Carry out a dry-weight determination on a representative portion of the test material to be analyses.
- Take a portion of wet test material, which does not contain more than 1g lipid or 8g of water.
- Weigh the test material with known moisture content in a 100ml centrifuge tube or appropriate glass jar.
- Add 18ml isopropanol and 20ml cyclohexane.
- Mix with Ultra Turrax for two minutes.
- Add W ml of water. W is calculated by :

$$W = 22 - \frac{\text{Sampleintake (g)} * \text{moisturecontent (\%)}}{100}$$

- Mix with Ultra Turrax for another minute.
- Separate the phases by centrifugation[2].
- Transfer as much as possible of the organic phase to an evaporation flask (by small pipette). Filtration is optional but makes the method more robust[3].

[1] When a centrifuge is not used, the phases may take time to separate and the interface is less sharp, which can result in a low recovery of the organic phase. A check should be made to determine whether > 80% of the organic phase has been recovered (18 ml). A third extraction is recommended in the case of a lower recovery.

[2] Some tissues, like liver extracts, form an emulsion which can be prevented by replacing the water by 1 M HClO4 to denature the proteins. The addition of NaCl may also help.

[3] In some cases the organic phase may contain some tissue particles when using the B & D Method. This also depends on the mixing method used (e.g. ultra sonic ). When this occurs the extract should be filtered by passing the extract through a glass column plugged with ca 2cm of cottonwool which has previously been extracted with solvent.

- Add 20 ml cyclohexane containing 13%(w/w) isopropanol and mix for one minute by Ultra Turrax.
- Centrifuge.
- Transfer the upper phase to the flask containing the first extract and evaporate the solvent.
- Quantitavely transfer the residue to a weighed wide-mouth cup by using a few ml of the cyclohexane/isopropanol mixture or diethylether.
- Evaporate in a moderately warm place to dryness (do not boil). The temperature used should be 5– 10 ˚C below the boiling point of the washing solvent. Evaporation may be assisted by a stream of nitrogen.
- Further dry the residue for one hour at 105 ˚C
- Weigh the residue and calculate the lipid content from the intake.