BS EN 16278:2012

BSI Standards Publication

Animal feeding stuffs — Determination of inorganic arsenic by hydride generation atomic absorption spectrometry (HG-AAS) after microwave extraction and separation by solid phase extraction (SPE)

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National foreword

This British Standard is the UK implementation of EN 16278:2012.

The UK participation in its preparation was entrusted to Technical Committee AW/10, Animal feeding stuffs.

A list of organizations represented on this committee can be obtained on request to its secretary.

ISBN 978 0 580 66999 6

ICS 65.120

This British Standard was published under the authority of the Standards Policy and Strategy Committee on 31 July 2012.

Amendments issued since publication

Date	Text affected

EUROPEAN STANDARD
NORME EUROPÉENNE
EUROPÄISCHE NORM

EN 16278

July 2012

ICS 65.120

English Version

Animal feeding stuffs - Determination of inorganic arsenic by hydride generation atomic absorption spectrometry (HG-AAS) after microwave extraction and separation by solid phase extraction (SPE)

Aliments des animaux - Dosage de l'arsenic inorganique par spectrométrie d'absorption atomique par génération d'hydrures après extraction par micro-ondes

Futtermittel - Bestimmung von anorganischem Arsen mit Atomabsorptionsspektrometrie-Hydridtechnik (HD-AAS) nach Mikrowellen-Extraktion und Trennung durch Festphasenextraktion (SPE)

This European Standard was approved by CEN on 17 May 2012.

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EUROPÄISCHES KOMITEE FÜR NORMUNG

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Ref. No. EN 16278:2012: E

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Foreword

This document (EN 16278:2012) has been prepared by Technical Committee CEN/TC 327 “Animal feeding stuffs”, the secretariat of which is held by NEN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by January 2013, and conflicting national standards shall be withdrawn at the latest by January 2013.

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1 Scope

This European Standard describes a procedure for the determination of inorganic arsenic in animal feeding stuffs of marine origin by Solid Phase Extraction (SPE) and Hydride Generation Atomic Absorption Spectrometry (HG-AAS). The method has been successfully tested in a collaborative trial with a working range from 0,19 mg/kg to 2,7 mg/kg (HORRATvalues < 2; HORRAT value is Horwitz –Ratio value). The LOQ of the method is usually approximately 0,1 mg/kg or lower.

2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

EN ISO 3696, *Water for analytical laboratory use — Specification and test methods (ISO 3696)*

EN ISO 6497, *Animal feeding stuffs — Sampling (ISO 6497)*

EN ISO 6498, *Animal feeding stuffs — Guidelines for sample preparation (ISO 6498)*

3 Principle

Inorganic arsenic consists of arsenite, As(III) and arsenate, As(V). This standard describes a method for the determination of inorganic arsenic (i.e. the sum of As(III) and As(V)). A representative test portion of the sample is treated with a diluted hydrochloric acid and hydrogen peroxide solution using microwave assisted heating. Hereby inorganic arsenic species are extracted and As(III) is oxidised to As(V). The inorganic arsenic is selectively separated from other arsenic compounds using solid phase extraction (SPE) and the concentration of inorganic arsenic is determined by HG-AAS in the SPE eluate. The gaseous hydride is transferred into a heated measuring cell (cuvette) by means of a carrier gas stream and decomposed.
Since arsenic (III) and arsenic (V) show a different sensitivity to the hydride technique, it is necessary to reduce arsenic (V) to arsenic (III) in order to avoid incorrect measurements.

WARNING — The use of this standard can involve hazardous materials, operations and equipment. This standard does not purport to address all the safety problems associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to its use.

4 Reagents

The concentration of arsenic in the reagents and water used shall be low enough not to affect the results of the determination. All reagents shall be of analytical grade or similar unless otherwise specified.

Use water conforming to grade 2 of EN ISO 3696.

4.1 Hydrochloric acid (HCl), mass fraction of ≥ 30 % of approximately ρ (HCl) ≥ 1,15 g/ml.

NOTE Only use hydrochloric acid available with high purity or which was cleaned by a sub-boiling distillation.

4.2 Diluted hydrochloric acid.

4.2.1 0,4 mol/l as eluent for the SPE.

Dilute e.g. 20 ml of hydrochloric acid (4.1) to 0,5 l with water.

4.2.2 0,055 mol/l for the extractant solution.

Dilute e.g. 5,8 ml of hydrochloric acid (4.1) to 1 l with water.

4.2.3 2,6 mol/l for sample pre-reduction.

Dilute e.g. 270 ml of hydrochloric acid (4.1) to 1 l with water.

4.2.4 4,7 mol/l for HG-AAS measurements.

Dilute e.g. 500 ml of hydrochloric acid (4.1) to 1 l with water.

4.3 Hydrogen peroxide (H_2O_2), mass fraction not less than 30 %.

4.4 Extractant solution, volume fraction of 3 % H_2O_2 in 0,055 mol/l HCl.

Dilute e.g. 20 ml hydrogen peroxide (4.3) to 200 ml with 0,055 mol/l hydrochloric acid (4.2.2).

4.5 Acetic acid, ρ (CH_3COOH) = 1,05 g/ml.

4.6 Diluted acetic acid, 0,5 mol/l for washing of SPE columns.

Dilute e.g. 15 ml of acetic acid (4.5) to 0,5 l with water.

4.7 Sodium hydroxide pellets, mass fraction of ≥ 98 %.

4.8 Sodium hydroxide, 0,1 mol/l for adjustment of pH.

Dilute 0.4 gram sodium hydroxide pellets (4.7) to 100 ml with water.

4.9 Ammonium carbonate, mass fraction of ≥ 99,999 %.

4.10 Ammonium carbonate buffer solution, 40 mmol/l for sample buffering.

Dissolve e.g. 0,384 g ammonium carbonate (4.9) in water and dilute to 100 ml with water.

4.11 Methanol (CH_3OH), HPLC grade, for conditioning of SPE sorbent.

4.12 Sodium borohydride, mass fraction of ≥ 96 %.

4.13 Sodium borohydride solution for hydride generation.

Dissolve e.g. 2,5 g of sodium hydroxide pellets (4.7) in a small amount of water; add 2,5 g of sodium borohydride (4.12) and dilute to 500 ml with water. Prepare a fresh solution daily prior to the hydride generation step and, when necessary, filter before use.

Commercially available sodium borohydride solutions may also be used.

NOTE The required concentrations may vary slightly with the system and the instructions of the relevant manufacturer shall therefore be observed.

WARNING — It is essential to observe the safety instructions for working with sodium borohydride. Sodium borohydride forms hydrogen with acids and this can result in an explosive air/hydrogen mixture. A permanent extraction system shall be provided at the point where the measurements are carried out.

4.14 L-Ascorbic acid, mass fraction of ≥ 99,7 %.

4.15 Potassium iodide, mass fraction of ≥ 99,5 %.

4.16 Pre-reduction solution for sample extract.

Dissolve 1,25 g of potassium iodide (4.15) and 1,25 g of L-ascorbic acid (4.14) up to 250 ml with 2,6 mol/l hydrochloric acid (4.2.3). Prepare a fresh solution on the day of analysis.

The required concentrations may vary slightly with the system and the instructions of the relevant manufacturer shall therefore be observed.

NOTE Addition of a volume fraction of 0,1 % silicone (4.21) to the solution can reduce the formation of foam in the gas-liquid separation system.

4.17 SPE equilibration solution.

Mix equal volumes (1+1) of 40 mmol/l ammonium carbonate solution (4.10) with extractant solution (4.4). 2 ml per SPE cartridge is needed.

4.18 Arsenic stock solution.

Commercially available standards with a mass concentration of 1 000 mg/l are recommended. Stock solutions in diluted nitric acid are preferred.

4.19 Diluted inorganic arsenic solutions.

Dilute the arsenic stock solution (4.18) by water in several steps, such as in the following example:

$$1\,000\ \text{mg/l} \xrightarrow[1/100]{} 10\ \text{mg/l} \xrightarrow[1/10]{} 1\ \text{mg/l} \xrightarrow[1/10]{} 0{,}1\ \text{mg/l}$$

4.20 Calibration solutions.

Prepare six external standards including a blank calibration solution that covers the linear range of the element to be determined by diluting the diluted inorganic arsenic solutions (4.19). The linear range is instrument dependent. Appropriate matching of the calibration solutions shall be performed (prereduction according to Table 2). It is important to adjust the acid concentration of the standards to the acid concentration of the samples.

The following calibration range is recommended: 0,25 µg/l to 8 µg/l. These calibration solutions should be prepared freshly before use.

NOTE An example on the production of calibration standard is given in Annex B, Table B.2.

4.21 Silicone antifoam agent ≥ 30 %.

5 Apparatus and equipment

To minimise contamination, all apparatus and equipment that come into direct contact with the sample and the solutions shall be carefully pre-treated. It is recommended to avoid the use of glassware, since this may cause contamination with arsenate [1].

5.1 Laboratory grinder, capable of grinding to a particle size of less than or equal to 0,5 mm.

5.2 Analytical balance, capable of weighing to an accuracy of 1 mg.

5.3 Microwave oven with closed vessels, capable of programming temperature.

5.4 pH indicator paper/sticks, for pH range 4 to 7.

5.5 Centrifuge with containers, capable of a minimum centrifuge speed of 4 000 rpm (2 100 *g*).

5.6 Solid phase extraction cartridges, with strong anion exchange stationary phase.

Strata SAX (Phenomenex), 500 mg / 6 ml have proven suitable for the present procedure. (Equivalent SPE columns with similar performance from other producers may also be used.)

NOTE Check the capacity of the SPE cartridges for retainment of inorganic arsenic.

5.7 Vacuum chamber or similar, for the elution process of the SPE.

5.8 One-mark volumetric flasks, of the following sizes: 100 ml, 150 ml, 250 ml, 500 ml, 1 000 ml.

5.9 Atomic absorption spectrometer, with measurement recording system, heated quartz cell and accessories for the hydride generation method.

5.10 Element-specific lamp, (electrode less discharge lamp (preferred) or hollow-cathode) for arsenic.

5.11 Automated flow system for hydride generation.

6 Procedure

6.1 General

IMPORTANT — The procedure is described in a flowchart (Annex B). Furthermore, Table B.1 provides information on which solutions and equipment are used for the different sections of the method: µ - wave, SPE and HG-AAS, respectively.

Sampling and preparation of the test sample is not part of this procedure. A recommended sampling method and method for sample preparation are given in EN ISO 6497 and EN ISO 6498.

6.2 Microwave assisted extraction

A test portion of approximately 0,2 g of the homogenised and ground (to a particle size of < 0,5 mm) test sample is weighed to an accuracy of 1 mg into a microwave oven vessel. 10,0 ml of the extractant solution (4.4) is added. The solution is then subjected to microwave-assisted heating using the temperature program outlined in Table 1.

Table 1 — Microwave oven programme

Step	Temperature	Time
1	90 °C	25 min
2	Cooling	7 min (time may vary; follow manufacturer’s instructions)

Following microwave treatment, the samples are allowed to cool to room temperature. The supernatant is then transferred to centrifuge containers and centrifuged for 10 min at ≥ 4 000 rpm (2 100 *g*).

NOTE The supernatant is used for subsequent SPE clean up and can be stored in clean containers at 4 °C for a maximum of three weeks until usage.

6.3 Solid phase extraction of inorganic arsenic

IMPORTANT — For adding liquids to the SPE cartridges, a low dropping speed is important. For adding a volume of 2 ml liquid, approximately 5 min elution time is recommended.

6.3.1 Sorbent conditioning of SPE cartridges

The SPE cartridges are conditioned with 2 ml methanol (4.11).

IMPORTANT — Let the SPE cartridges run dry.

6.3.2 Sorbent equilibration of SPE cartridges

The SPE cartridges are conditioned with 2 ml SPE sorbent equilibration solution (4.17).

IMPORTANT — Let the SPE cartridges run dry.

6.3.3 Buffering of sample solution

3 ml of the sample supernatant (6.2) is mixed with 3 ml 40 mmol/l ammonium carbonate buffer solution (4.10). Ensure the pH of the solution is 6,5 ± 1 with pH indicator paper/sticks (5.4). Adjust if necessary with diluted acetic acid (4.6) or diluted sodium hydroxide (4.8).

Centrifuge the buffered sample solution at > 4 000 rpm (2 100 *g*) for at least 10 min to remove particles.

6.3.4 Loading of sample solution

4 ml of the centrifuged buffered sample solution (6.3.3) is loaded to the SPE cartridges.

IMPORTANT — Let the SPE cartridges run dry.

6.3.5 Washing of SPE cartridges

Wash the SPE cartridges with 3 ml 0,5 mol/l acetic acid (4.6).

IMPORTANT — Let the SPE cartridges run dry, using vacuum (50 kPa to 70 kPa) for at least 5 min before the final elution step (6.3.6).

6.3.6 Elution of SPE cartridges

The retained As(V) on the SPE cartridge is eluted with 1,25 ml of 0,4 mol/l hydrochloric acid (4.2.1).

IMPORTANT — Collect all sample solvent in suitable plastic containers by applying suitable vacuum (usually in the range of 50 kPa to 70 kPa) for at least 5 min.

NOTE The eluate (6.3.6) can usually be stored at maximum 4 °C for a maximum of three days until analysis.

6.4 Determination of inorganic arsenic

6.4.1 General

The inorganic arsenic is determined as the total amount of arsenic in the SPE eluate. Carry out pre-reductions of test solutions (6.3.6) and calibration solutions (4.20) prior to the determination.

6.4.2 Pre-reduction

This step is dependent on the hydride system used. Therefore, please follow the optimised hydride generation procedure of the equipment used and note that it may be necessary to use larger or smaller volumes than described below (Table 2).

Table 2 — Example of a procedure for the pre-reduction of test solutions prior to HG-AAS measurement

Aliquot of test solution (6.3.6)	1 ml
Potassium iodide/ascorbic acid solution with hydrochloric acid (4.16)	Add 7 ml and mix thoroughly
Incubation	60 min at room temperature
2,6 mol/l hydrochloric acid (4.2.3)	Add 6 ml and mix thoroughly
Incubation	60 min at room temperature

All samples including the calibration solutions should be treated in the same way. The hydrochloric acid (4.2.3) and reducing-agent (4.16) concentrations shall be the same in all the test solutions (see Annex B, Table B.2 for an example).

6.4.3 Settings of the Atomic absorption spectrometer (HG-AAS procedure)

To devise a test schedule, first adjust the apparatus as specified in the operating manual of the manufacturer. Then optimise the settings, paying particular attention to gas flow times and the amount of sodium borohydride introduced. Typical settings are listed in Table 3.

Table 3 — Typical settings of HG-AAS for arsenic measurement

Temperature of the cell	900 °C
Wave length	193,7 nm
Slit width	0,5 nm
Signal processing	Peak height with background correction
Measurement time	4,0 s

6.4.4 HG-AAS determination

The pre-reduced test solutions and pre-reduced calibration solutions are measured directly with an atomic absorption spectrometer with electrically heated quartz cell coupled to an automated flow system (5.11). The apparatus should be programmed to mix appropriate amounts of test- or calibration solution (6.4.2), diluted hydrochloric acid (4.2.4) and borohydride solution (4.13). The resulting gas/liquid mixture is separated by an argon-flow separator. The argon stream separates and transports the arsenic hydrides to the quartz cell for atomisation reaction and measuring the atomic absorption of arsenic.

The pre-reduced calibration solutions are measured first; then the test solutions are measured. Reanalyse the calibration solutions at the end of each analytical series.

As an analytical control, reference samples having reliable known inorganic arsenic contents can be analysed parallel with all the series of samples analysed, the reference samples being subjected to all the steps in the method starting from microwave extraction. Blank solutions prepared by subjecting them to all the steps in the method shall also be determined.

7 Calculation

In general, the calibration curve and the element concentration of the test solution are calculated by the AAS system software.

The mass fraction *w* in milligram inorganic arsenic per kilogram sample is calculated as follows:

$$w = \frac{c_t \times V_1 \times V_3 \times V_5 \times m_{extr}}{V_2 \times V_4 \times V_6 \times \rho \times m_t \times 1000} \tag{1}$$

where (typical values in parenthesis)

c_t	is the concentration of arsenic in the test solution, in µg/l;
V_1	is the total volume of the pre-reduction solution, in ml (14 ml);
V_2	is the sample volume for pre-reduction step, in ml, (1 ml);
V_3	is the volume from SPE elution, in ml, (1,25 ml);
V_4	is the volume put on the SPE, in ml, (4 ml);
V_5	is the total volume of buffered sample solution, in ml, (6 ml);
V_6	is the volume of supernatant for the buffered sample solution, in ml, (3 ml);
m_{extr}	is the mass of extractant solution after microwave extraction step, in g, (10 g);
ρ	is the density of extractant solution (g/ml), (1 g/ml);
m_t	is the mass of test portion, in g.

If the typical values in parenthesis are used, Formula (1) can be simplified as follows:

Simplified formula: $$w = \frac{c_t \times 0{,}0875}{m_t} \tag{2}$$

If necessary, subtract the concentration of inorganic arsenic in the reagent blank from c_t.

8 Precision

8.1 General

An interlaboratory comparison was organized by the National Food Institute, Technical University of Denmark (DTU Food) in collaboration with the Institute for Reference Materials and Measurements (IRMM) in 2010. The results of the main method protocol are given in Annex A. Further details regarding the outcome of the collaborative study can be found in the final report [2].

Details of an interlaboratory test on the precision of the method are summarized in Annex A. The values derived from this interlaboratory test may not be applicable to concentration ranges and matrices other than those given in Annex A.

8.2 Repeatability

The absolute difference between two independent single test results, obtained with the same method on identical test material in the same laboratory by the same operator using the same equipment within a short interval of time, will in no more than 5 % of the cases exceed the values of r (repeatability limit) given in Table 4.

8.3 Reproducibility

The absolute difference between two single test results, obtained with the same method on identical test material in different laboratories by different operators using different equipment, will in no more than 5 % of the cases exceed the values of R (reproducibility limit) given in Table 4.

Table 4 — Precision data

	Fish feed	**Fish meal**	**Fish meal**	**Fish fillet**	**Fish meal**	**Lobster Hepatopancreas**
Mean, mg/kg	0,725	0,194	1,083	2,705	0,439	0,555
r, mg/kg	0,138	0,042	0,307	0,490	0,068	0,272
R, mg/kg	0,315	0,179	0,363	1,397	0,189	0,467
NOTE r is the repeatability limit; R is the reproducibility limit.						

9 Test report

The test report shall specify at least the following:

a) information necessary for the complete identification of the sample;

b) test method used with reference to this European standard;

c) test results obtained and the units in which they are specified;

d) date of sampling and sampling procedure (if known);

e) date when the analysis was finished;

f) all operating details not specified in this document, or regarded as optional, together with details of any incidents that occurred when performing the method which may have influenced the test result(s).

Annex A
(informative)

Results of the interlaboratory tests

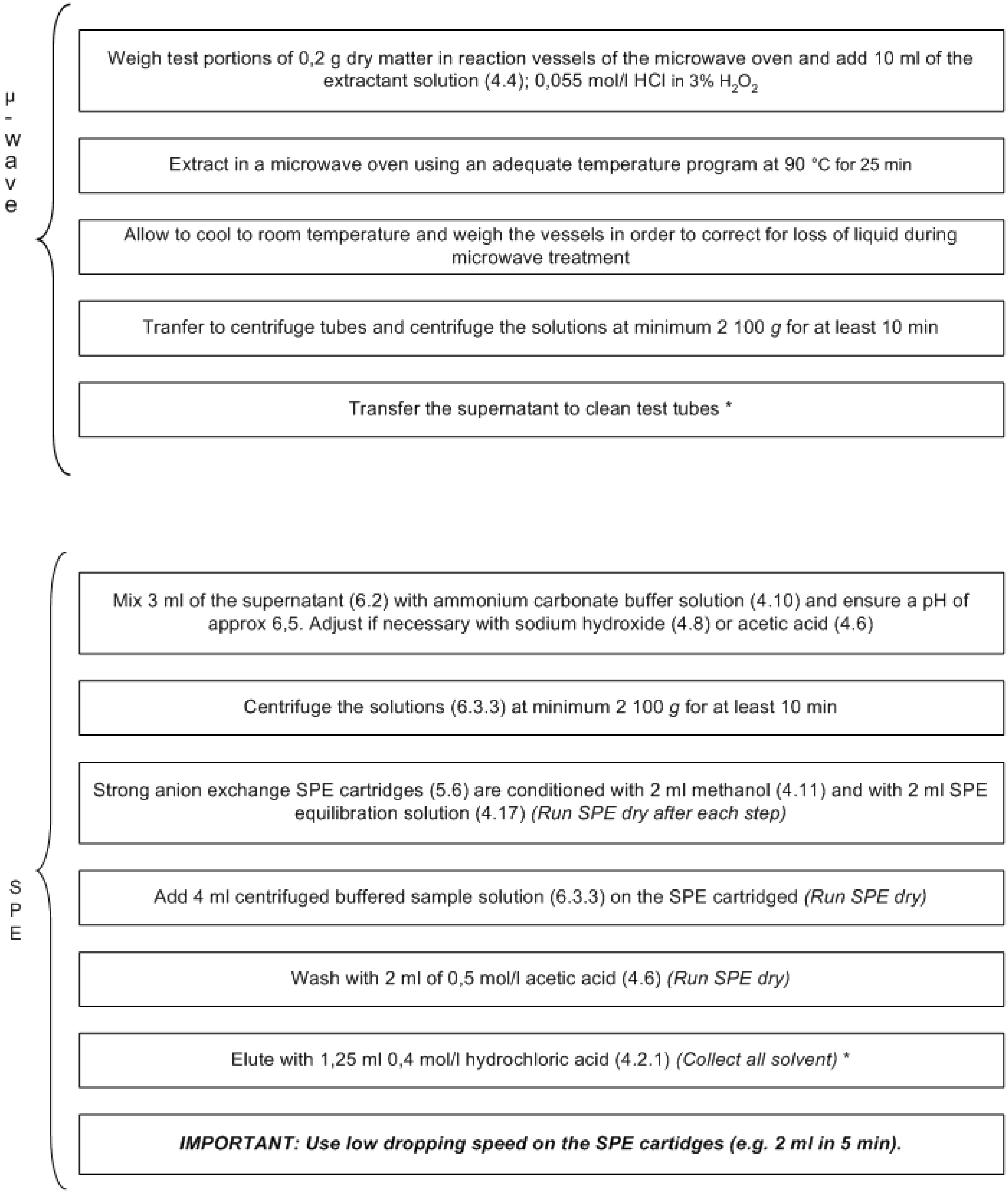
Table A.1 — Precision data

Matrix	Units	Fish feed	Fish meal	Fish meal	Fish fillet	Fish meal	Control Sample
Number of labs		9	9	9	9	9	9
Number of outlier labs		0	1	0	1	1	0
Number of valid labs		9	8	9	8	8	9
Remaining data after outlier elimination		31	24	32	28	26	31
Outliers	%	3,1	14,3	0,0	12,5	13,3	0,0
Overall mean X_{obs}	mg/kg	0,725	0,194	1,083	2,705	0,439	0,555
s_r	mg/kg	0,049	0,015	0,110	0,175	0,024	0,097
CV_r	%	6,8	7,7	10,1	6,5	5,5	17,5
r	mg/kg	0,138	0,042	0,307	0,490	0,068	0,272
s_R	mg/kg	0,113	0,064	0,130	0,499	0,068	0,167
CV_R	%	15,5	33,0	12,0	18,4	15,4	30,1
R	mg/kg	0,315	0,179	0,363	1,397	0,189	0,467
HORRAT value		0,93	1,61	0,76	1,30	0,85	1,7

NOTE r is the repeatability limit; S_r is the repeatability standard deviation; CV_r is the repeatability variation coefficient; HORRAT value is Horwitz –Ratio value; R is the reproducibility limit; S_R is the reproducibility standard deviation; CV_R is the reproducibility variation coefficient.

Annex B
(informative)

Flowchart Animal feeding stuffs — Determination of inorganic arsenic by hydride generation atomic absorption spectrometry (HG-AAS) after microwave extraction and separation by solid phase extraction (SPE)



Weigh test portions of 0,2 g dry matter in reaction vessels of the microwave oven and add 10 ml of the extractant solution (4.4); 0,055 mol/l HCl in 3% H_2O_2

Extract in a microwave oven using an adequate temperature program at 90 °C for 25 min

Allow to cool to room temperature and weigh the vessels in order to correct for loss of liquid during microwave treatment

Tranfer to centrifuge tubes and centrifuge the solutions at minimum 2 100 *g* for at least 10 min

Transfer the supernatant to clean test tubes *

Mix 3 ml of the supernatant (6.2) with ammonium carbonate buffer solution (4.10) and ensure a pH of approx 6,5. Adjust if necessary with sodium hydroxide (4.8) or acetic acid (4.6)

Centrifuge the solutions (6.3.3) at minimum 2 100 *g* for at least 10 min

Strong anion exchange SPE cartridges (5.6) are conditioned with 2 ml methanol (4.11) and with 2 ml SPE equilibration solution (4.17) *(Run SPE dry after each step)*

Add 4 ml centrifuged buffered sample solution (6.3.3) on the SPE cartridged *(Run SPE dry)*

Wash with 2 ml of 0,5 mol/l acetic acid (4.6) *(Run SPE dry)*

Elute with 1,25 ml 0,4 mol/l hydrochloric acid (4.2.1) *(Collect all solvent)* *

IMPORTANT: Use low dropping speed on the SPE cartidges (e.g. 2 ml in 5 min).

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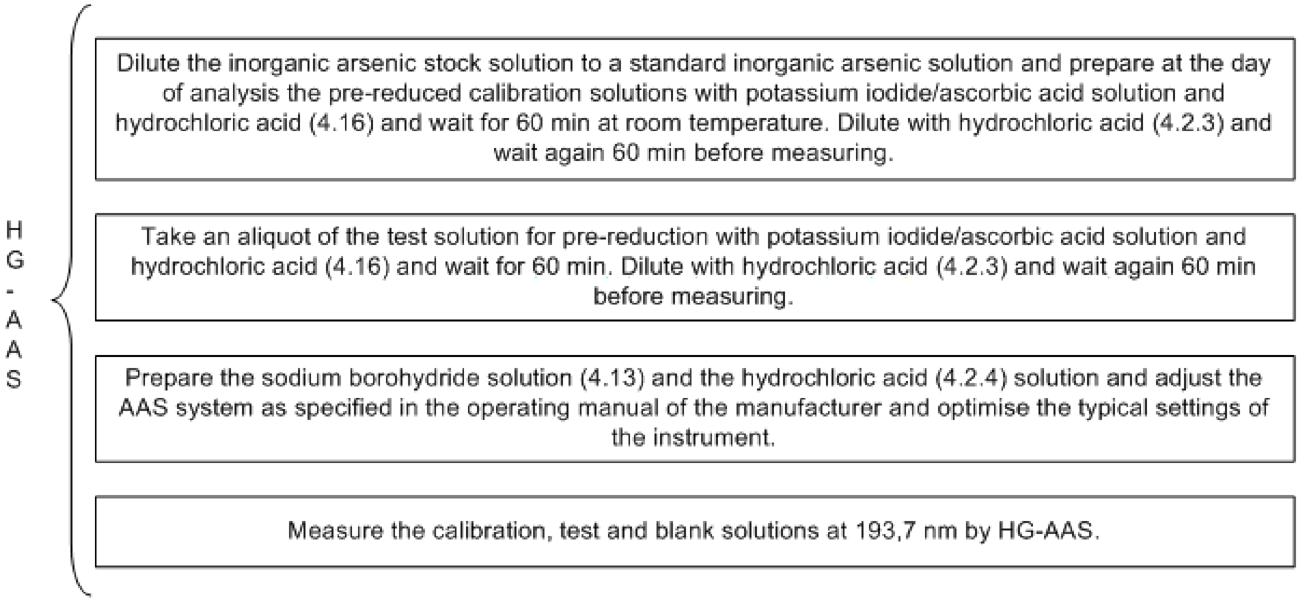
* The solutions can be stored (at 4 °C) for later continuation of the analysis.



Dilute the inorganic arsenic stock solution to a standard inorganic arsenic solution and prepare at the day of analysis the pre-reduced calibration solutions with potassium iodide/ascorbic acid solution and hydrochloric acid (4.16) and wait for 60 min at room temperature. Dilute with hydrochloric acid (4.2.3) and wait again 60 min before measuring.

Take an aliquot of the test solution for pre-reduction with potassium iodide/ascorbic acid solution and hydrochloric acid (4.16) and wait for 60 min. Dilute with hydrochloric acid (4.2.3) and wait again 60 min before measuring.

Prepare the sodium borohydride solution (4.13) and the hydrochloric acid (4.2.4) solution and adjust the AAS system as specified in the operating manual of the manufacturer and optimise the typical settings of the instrument.

Measure the calibration, test and blank solutions at 193,7 nm by HG-AAS.

The following Table B.1 gives an overview of which chemicals, solutions and equipment are used for the three different steps in the analytical procedure

Table B.1 — Solutions and equipment

Step	**Chemicals and solutions**	**Equipment**
µ-wave	Water ≥ 30 % Hydrochloric acid (HCl) (4.1) 0,055 mol/l HCl (4.2.2) ≥ 30 % Hydrogen peroxide (H_2O_2) (4.3) 3 % H_2O_2, 0,055 mol/l HCl (4.4) for sample extraction	Microwave oven (5.3) Closed vessels (5.3) Balance (5.2) Centrifuge (5.5) Centrifuge containers (5.5) Volumetric flasks (5.8)
SPE	100 % methanol (4.11) for conditioning of SPE columns Water ≥ 99,999 % ammonium carbonate (4.9) 40 mmol/l ammonium carbonate (4.10) ≥ 30 % Hydrogen peroxide (H_2O_2) (4.3) 3 % H_2O_2, 0,055 mol/l HCl (4.4) prepared earlier for the µ-wave step SPE equilibration solution (4.17) Acetic acid (4.5) 0,5 mol/l acetic acid (4.6) for washing of SPE columns ≥ 30 % HCl (4.1) 0,4 mol/l HCl (4.2.1) for sample elution of SPE columns	SPE cartridges (5.6) pH sticks (5.4) Centrifuge containers (5.5) Volumetric flasks (5.8)

Step	Chemicals and solutions	Equipment
HG-AAS	Water ≥ 99,7 % L-Ascorbic acid (4.14) ≥ 99,5 % Potassium iodide (4.15) ≥ 30 % HCl (4.1) 2,6 mol/l HCl (4.2.3) Antifoam silicone oil (4.21) Pre-reduction solution (4.16), fresh 4,7 mol/l HCl (4.2.4) ≥ 96 % Sodium borohydride (4.12) ≥ 98 % Sodium hydroxide pellets (4.7) Sodium borohydride solution (4.13), fresh Arsenic stock solution (4.18) Diluted arsenic solutions (4.19) Calibration solutions (4.20)	HG-AAS instrument (5.9) Volumetric flasks (5.8)

The following Table B.2 gives an example of how to prepare a range of calibration standards. Follow the instructions in 6.4.2.

Table B.2 — Example of preparation of calibration solutions

AAS standard µg/l	Inorganic As solution in water (4.19) mg/l	Inorganic As solution µl	Pre-reduction solution (4.16) wait 60 min ml	2,6 mol/l HCl (4.2.3) wait 60 min ml
0	-	0	15	15
0,25	0,1	100	20	20
0,50	0,1	200	20	20
0,99	0,1	400	20	20
2,49	1,0	100	20	20
4,98	1,0	200	20	20
8,06	1,0	325	20	20

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[3] ISO 5725-1, *Accuracy (trueness and precision) of measurement methods and results — Part 1: General principles and definitions*

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