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IMEP-116: Determination of total cadmium, lead, arsenic, mercury and inorganic arsenic in mushrooms

Interlaboratory Comparison Report

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IMEP-116: Determination of total cadmium, lead, arsenic, mercury and inorganic arsenic in mushrooms

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Executive summary

The Institute for Reference Materials and Measurements (IRMM) of the Joint Research Centre (JRC), a Directorate-General of the European Commission, operates the European Union Reference Laboratory for Heavy Metals in Feed and Food (EURL-HM). One of its core tasks is to organise proficiency tests (PTs) among appointed National Reference Laboratories (NRLs), in support to European Union (EU) policies. This report presents the results of a PT, IMEP-116, on the determination of total cadmium, lead, arsenic and mercury and of inorganic arsenic in mushrooms. The exercise was organised in support to the European Commission Regulation (EC) No 1881/2006, which sets the maximum levels for certain contaminants in foodstuffs.

Thirty eight participants from twenty six countries registered to the exercise. Only one participant did not report results.

The test item used was a blend of mushrooms of the variety shiitake (*Lentinula edodes*). Five laboratories with demonstrated measurement capability in the field provided results to establish the assigned values (X_{ref}). The standard uncertainties associated to the assigned values (u_{ref}) were calculated according to ISO/IEC Guide 98:2008 (GUM) by combining the uncertainty of the characterisation (u_{char}) with a contribution for homogeneity (u_{bb}) and for stability (u_{st}). u_{char} was calculated following ISO 13528:2005.

Laboratory results were rated with *z*- and zeta (*ζ*-) scores in accordance with ISO 13528 and ISO 17043:2010. The standard deviation for the proficiency assessment, σ_p, for total Pb (20 %) and inorganic arsenic (19 %) was calculated using the Horwitz equation as modified by Thompson. For the rest of the measurands, σ_p was set by the advisory board of this PT to 15 % for total As and Hg and to 10 % for total Cd, on the basis of previous participant's performance on similar measurands.

The percentage of satisfactory z-scores ranged from 81 % (inorganic arsenic) to 97 % (total cadmium). Therefore, the outcome of the exercise shows an appropriate performance for EU National Reference Laboratories assuring compliance towards the EU legislation related to the determination of the investigated food contaminants.

1   Introduction

Mushrooms are excellent sources of nutrients (proteins, fibre, vitamins and essential minerals). However, edible portions of mushrooms are also known to accumulate high levels of heavy metals such as cadmium, lead, arsenic and mercury from the soil [1].

To protect consumers from toxic effects, maximum levels for heavy metals in mushrooms have been laid down in Regulation (EC) No 1881/2006 [2] and its amendments [3, 4].

The Directorate General for Health and Consumers (DG SANCO) of the European Commission, requested the EURL-HM to test the analytical capabilities of the NRLs regarding the determination of heavy metals in mushrooms. Therefore, the EURL-HM organised a PT, IMEP-116, for the determination of total Cd, Pb, As, Hg and inorganic As in shiitake (*Lentinula edodes*), the mushroom most cultivated and consumed worldwide, thus deserving a particular attention.

IMEP-116 was coordinated by the International Measurement Evaluation Programme (IMEP), who organised in parallel the PT IMEP-39 to assess the performance of European and non-European food control laboratories (FCL), using the same test item and the same criteria for performance evaluation.

This report summarises and evaluates the outcome of IMEP-116, but does not discuss the outcome of IMEP-39.

2   Scope and aim

As stated in Regulation (EC) No 882/2004 [5] one of the core duties of the EURLs is to organise interlaboratory comparisons for the benefit of staff from National Reference Laboratories. The scope of this proficiency test was to test the competence of the appointed NRLs to determine the total Cd, Pb, As, Hg and inorganic As mass fractions in mushrooms.

The assessment of the measurement results was undertaken on the basis of requirements laid down in European legislation [2], and followed the administrative and logistic procedures of ISO 17043:2010. The Institute of Reference Materials and Measurements of the European Commission Directorate the Joint Research Centre (EC-JRC) is accredited according to ISO 17043:2010 [6].

3   Set-up of the exercise

3.1   Time frame

The exercise was agreed upon by the NRL network at the seventh EURL-HM workshop held in September 2012. Invitation letters for participation were sent to the

NRLs on 7 March 2013 (Annex 1). The PT exercise was announced via the IMEP web page on 13 March 2013 (Annex 2).

Registration was opened till 30 April 2013. The deadline for reporting results was the 22 June 2013. Dispatch was followed by the PT coordinator using the messenger's parcel tracking system on the internet.

3.2   Distribution

Test items were dispatched on 15 May 2013. Each participant received one package containing:

- One bottle containing approximately 2.5 g of the test item,

- The "*Sample accompanying letter*" (Annex 3),

- A *"Confirmation of Receipt"* form (Annex 4).

3.3   Instructions to participants

Participants were asked to perform two or three independent measurements, correct their measurements for recovery and for the moisture content (protocol provided in the sample accompanying letter) and report their calculated mean (expressed on a dry mass) and its associated measurement uncertainty (u_{lab}).

Participants received an individual code to access the online reporting interface, to report their measurement results and to complete the related questionnaire. The questionnaire was used to extract all relevant information related to measurements and laboratories (Annex 5).

Participants were asked to follow their routine procedures for the analysis and to report results in the same way (e.g. number of significant figures) as they would report to their customers.

4   Proficiency test item

4.1   Preparation

Approximately 5 kg of fresh shiitake mushrooms (*Lentinula edodes*), was screened for the measurands covered in IMEP-116 and provided by the University of Barcelona (Spain). Fresh mushrooms were hand-cleaned for soil, moss, etc. The end of the stalk that had been in contact with soil was cut off using a stainless steel knife. Mushrooms were cut into pieces, which were air dried in a batch-type drying chamber at room temperature for 24 hours and dried in an oven at 40 °C for 24-48 hours. Dried mushrooms were minced using a commercial stainless steel mincer (Multiquick 5 Hand Processor, Braun), completely homogenized, packaged and dispatched immediately to IRMM under refrigerated conditions.

Once received, the material was stored at -20 °C until processing. At the time of processing the mushrooms were cut frozen in smaller pieces using an UMC-12 model cutter/mixer from Stephan (Hameln, DE).

The material was freeze dried in two cycles using a freeze dryer from Martin Christ model Epsilon 2-10D (Osterode, DE). Five trays were filled with about 500 g each of pre-cut mushrooms per cycle. In total 5.27 kg were dried, giving 570 g of dried mushroom, corresponding to a mass loss of about 89 %.

Dried mushrooms were cryogenically milled using a Palla VM-KT vibrating mill from Humboldt-Wedag (Köln, DE). All grinding elements in this system were made of high purity titanium to avoid contamination of test material. After milling, this material was sieved over a 250 µm stainless steel sieve resulting in 522 g available for final mixing and homogenisation. Mixing was performed in a Dynamix CM-200 (WAB, Basel, CH). The Karl Fischer titration and laser diffraction analysis indicate that the material had a water content of 4 % (m/m) with a top particle size below 200 µm.

Finally, portions of 2.5 g were filled using an automatic filling machine (Allfill, Sandy, UK) into 20 ml amber glass acid-washed vials. The vials were closed with acid washed inserts and aluminium caps.

Each vial was uniquely identified (labelled following the IMEP procedures) including a unique number and the name of the PT exercise.

4.2   Homogeneity and stability studies

The homogeneity and stability studies were performed by ALS Scandinavia AB (Sweden) using inductively coupled plasma sector field mass spectrometry (ICP-SFMS) after sample digestion with a mixture of HNO_3/HF. Homogeneity was evaluated according to ISO 13528 [7]. The material proved to be adequately homogeneous for the total mass fraction of As, Cd, Pb and Hg.

The stability study was conducted following an isochronous experimental design [8, 9]. The material proved to be adequately stable for the eight weeks that elapsed between the dispatch of the samples and the deadline for submission of results and for all the four investigated total mass fractions (As, Cd, Pb and Hg).

It was assumed, on the basis of previous experience (IMEP-107), that, if adequately homogeneous and stable for the total mass fraction of As, it should also be for the inorganic form of that element.

The contributions due to homogeneity (u_{bb}) and to stability (u_{st}) to the uncertainty of the assigned value (u_{ref}) were calculated using SoftCRM [10]. For iAs identical contributions were calculated using the same percentage (of the mean value) as estimated for the total mass fraction of As.

The analytical results and the statistical evaluation of the homogeneity and stability studies are provided in Annex 6.

5 Reference values and their uncertainties

5.1 Assigned value X_{ref}

Five expert laboratories (certifiers) analysed the test item in order to determine the assigned value:

- *Federal Institute for Materials Research and Testing (BAM), Germany*

- *Laboratorio de Salud Pública de Alicante (LSPA), Spain*

- *Karl-Franzens-Universität Graz (KFUG), Austria*

- *University of Barcelona, Faculty of Chemistry (UBFC), Spain*

- *Instituto de Agroquímica y Tecnología de los Alimentos, Consejo Superior de Investigaciones Científicas (CSIC), Spain*

Expert laboratories were asked to use the method of their choice and no further requirements were imposed regarding methodology. Expert laboratories were also asked to report their results together with the measurement uncertainty and with a clear and detailed description on how uncertainty was estimated.

The mean of the independent means provided by the certifiers was used to derive the assigned values (X_{ref}) for this PT according to ISO Guide 35:2006 [11].

Table 1 summarises the sample preparation and digestion procedures and details related to the analytical method used by the certifiers.

Table 1 – *Sample treatment, digestion procedures and analytical methods used by the certifiers*

Certifier	Sample treatment / digestion / analytical method	Technique
BAM	**Total As, Cd and Pb**: 0.25 g of sample. Microwave-assisted digestion. 6 mL of HNO_3 (sub-boiling) in an Ultra Clave III. Power 1000 W, ramp 20 min. hold 30 min. Digestion temperature 250 °C at 100 bar. ICP equipped with a collision cell. Argon + helium as collision gas. Multi-point calibration from 0 - 10 µg L^{-1} (5 points) for total As and Pb, 0 – 25 µg L^{-1} for Cd.	ICP-MS
BAM	**Total Hg**: 0.25 g o f sample. Microwave-assisted digestion. 6 mL of HNO_3 (sub-boiling) in an Ultra Clave III. Power 1000 W, ramp 20 min. hold 30 min. Digestion temperature: 250 °C at 100 bar. CV-AFS, amalgamation mode (gold trap). Argon as gas. Multi-point calibration from 0-125 µg L^{-1} (5 points).	CV-AFS
BAM	**Total Hg**: 0.12 g of sample. Solid sampling cold-vapour AAS, combustion + amalgamation (gold trap). Advanced elementar mercury analyser (AMA-254) at the wavelength of 253.7 nm. Oxygen as gas mode. Multi-point calibration from 0.5 – 36 ng (9 points) and from 40 to 500 ng (9 points).	Elemental Hg analyser
LSPA	**Total As, Cd, Pb**: The digestion of samples was carried out using a microwave digestion system, Ethos one (Milestone Inc., Shelton, USA), equipped with the Q-20 Quartz Rotor Ultratrace Analysis (20 mL quartz tubes, 250 ºC and 40 bars operating parameters). A unique sample digestion procedure was applied to all samples and analytes. 0.25 g of sample was weighted in quartz digestion vessels and 5 mL of HNO_3:H_2O 1:1 were added in a fume hood. The mixture was leaved to react over an hour approximately until finishing the gas generation process. Analysis were performed on an ELAN DRC II ICP-MS (PerkinElmer, Inc., Shelton, USA) equipped with a PFA standard nebulizer and a peltier cooled baffled glass cyclonic spray chamber (both from Elemental Scientific, Omaha, USA). Multi-element standard solutions were used for external calibration. Six standards in 2 % (w/w) HNO_3 matrix for As, Cd and Pb were prepared at levels ranging from 0.1 to 50 µg L^{-1}. The calibration curve was drawn from six points, including the calibration blank and there was applied a weighted linear regression approach with internal standardization.	ICP-MS
LSPA	**Total Hg**: 40 mg of sample was weighted directly in quartz samples boats and placed in the mercury analizer. To prevent explosions inside the catalizer, 500 µL of ultra-pure water were added in the quartz boats together with the samples. At least 2 quality control samples (CRM) were analysed in each sequence.	Elemental Hg analyser
KFUG	**Total As**: A portion of the powdered samples (about 250 mg weighed with a precision of 0.1 mg) was weighed directly into 12 mL quartz tubes, and concentrated nitric acid (2 mL) and H_2O (2 mL) were added. The tubes were transferred to a Teflon® rack of the Ultraclave microwave system (MLS GmbH, Leutkirch, Germany) and covered with Teflon® caps. After closing the system, an argon pressure of 4 x 106 Pa was applied and the mixture was heated to 250 °C for 30 minutes before being allowed to cool to room temperature. After mineralization, the samples were transferred to 15 mL polypropylene tubes (Greiner, Bio-one, Frickenhausen, Germany) and diluted with water to 9 mL (based on mass). Finally 1 mL of a solution containing 50 % methanol (to enhance the arsenic response) and 100 µg·L^{-1} each of Ge and In as internal standards were added to all digested samples giving a final concentration of 5 % methanol and 10 µg·L^{-1} of Ge and In. All standards for total arsenic determinations were prepared with 20% (v/v) of concentrated nitric acid and also 5% methanol for matrix matching with the digested samples. The arsenic concentrations in the digests were determined by ICP-MS using helium as collision cell gas.	ICP-MS
KFUG	**Inorganic As**: About 0.5 g of powder was weighed with a precision of 0.1 mg into 50 mL polypropylene tubes, and a solution (10 mL) of 20 mmol·L^{-1} trifluoracetic acid containing 50 µL of a 30 % H_2O_2 solution was added. Samples were extracted with a GFL-1083 shaking water bath (Gesellschaft für Labortechnik, Burkwedel, Germany) at 95 °C for 60 minutes. After cooling to room temperature the extracts were centrifuged for 15 min at 4700 g. An aliquot of 1 mL was transferred to Eppendorf vials and centrifuged for 15 min at 8900 g. The supernatant was used directly for HPLC-ICP-MS analysis.	HPLC-ICP-MS

Certifier	Sample treatment / digestion / analytical method	Technique
CSIC	**Inorganic As**: 0.5-1 g of sample. Concentrated HCL is added and water. Reducing agent (2 mL of HBr and 1 mL of hydrazine sulphate) is added. 10 mL of $CHCl_3$. Agitate and separate the phases. Repeat the extraction 3 times. iAs is back-extracted with 10 mL of HCl. 2.5 mL of ashing aid suspension (20 % w/v $Mg(NO_3).6H_2O$ and 2 % w/v MgO) and 10 mL HNO_3 is added. Evaporated to dryness in a sand bath and place at a muffle at 150 °C. Increase the temperature to 425 ± 25 °C for 12 H. The white ash is dissolved in 6 mol L^{-1} HCl and reduced with pre-reducing solution (5 % w/v KI and 5 % w/v ascorbic acid). After 30 min, filter through Whatman N° 1 and dilute with 6 mol L^{-1} HCl. Samples are analysed by flow injection-hydride generation AAS.	FI-HG-AAS
UBFC	**Inorganic As**: A microwave digestion system, Ethos Touch Control (Milestone, Gomensoro, Barcelona, Spain), with a microwave power of 1000 W and temperature control, was used for extraction procedure. An Agilent 7500ce ICPMS was coupled to an Agilent 1200 LC quaternary pump to determine inorganic arsenic content. The analytical columns Hamilton PRP-X100 (250x4.1 mm, 10 µm, Hamilton, USA) and Zorbax-SCX300 (250x 4.6 mm, 5 µm, Agilent) were protected by guard columns filled with the corresponding stationary phases. The outlet of the LC column was connected via PEEK capillary tubing to the nebuliser (BURGENER Ari Mist HP type) of the ICP-MS system, which was the arsenic-selective detector. 0.25-g aliquots of the test material and the CRMs were weighed in PTFE vessels and then extracted by adding 10 mL of 0.2 % (w/v) HNO_3 and 1 % (w/v) H_2O_2 solution in a microwave digestion system. The temperature was raised first to 55 °C (and held for 10 min) then to 75 °C (and held for 10 min) and finally the digest was taken up to 95 °C and maintained for 30 min. Samples were cooled to room temperature and centrifuged at 3500 rpm for 12 min. The supernatant was filtered through PET filters (pore size 0.45 µm).	HPLC-ICP-MS

5.2 Associated uncertainty u_{ref}

The standard uncertainties associated to the assigned values (u_{ref}) were calculated according to ISO/IEC Guide 98:2008 (GUM) [12] by combining the uncertainty of the characterisation (u_{char}) with a contribution for homogeneity (u_{bb}) and for stability (u_{st}), according to equation 1:

$$u_{ref} = \sqrt{u_{char}^2 + u_{bb}^2 + u_{st}^2}$$
$$\text{Eq. 1}$$

u_{char} was calculated combining the standard uncertainties reported by the expert laboratories (u_i) according to ISO 13529:2005 [7] (equation 2):

$$u_{char} = \frac{1.25}{p} \sqrt{\sum_1^p u_i^2}$$
$$\text{Eq. 2}$$

Where p refers to the number of expert laboratories used to assign the reference value.

Table 2 presents the results reported by the expert laboratories, standard uncertainty contributions, the reference values (X_{ref}, u_{ref} and U_{ref}) and the standard deviation for the PT assessment σ_p.

Table 2 – *Reported values by the expert laboratories (X_n), their uncertainty contributions (U_n), assigned value, standard and combined uncertainties u_{ref} (in mg kg^{-1})*

	Certifier	Total As	Total Cd	Total Pb	Total Hg	iAs
$X_n \pm U_n$	*BAM*	0.638 ± 0.026	4.42 ± 0.19	0.274 ± 0.019	0.0782 ± 0.0032 0.0781 ± 0.007	
	LSPA	0.61 ± 0.06	3.99 ± 0.44	0.260 ± 0.016	0.072 ± 0.007	
	KFUG	0.69 ± 0.05				0.330 ± 0.014
	CSIC					0.286 ± 0.037
	UBFC					0.348 ± 0.026
X_{ref}		**0.646**	**4.21**	**0.267**	**0.076**	**0.321**
u_{char}		0.017	0.15	0.008	0.002	0.010
u_{bb}		0.007	0.04	0.009	0.002	0.004
u_{st}		0.015	0.06	0.010	0.002	0.007
u_{ref}		0.024	0.17	0.016	0.004	0.013
U_{ref} (k=2)		**0.048**	**0.33**	**0.031**	**0.007**	**0.026**
σ_p		0.10	0.42	0.05	0.011	0.06
σ_p (%)		**15%**	**10%**	**20%**	**15%**	**19%**

5.3　Standard deviation for the proficiency test assessment σ_p

The standard deviation for the proficiency assessment (σ_p) for total Pb (20 %) and inorganic arsenic (19 %) were calculated using the Horwitz equation as modified by Thompson [13]. For the rest of the measurands σ_p was set by the advisory board of this PT to 15 % for total As and Hg and to 10 % for total Cd, on the basis of previous participant's performance on similar measurands.

6　Evaluation of results

6.1　Scores and evaluation criteria

Individual laboratory performance is expressed in terms of *z-* and *ζ-scores* in accordance with ISO 13528 [7]:

$$z = \frac{x_{lab} - X_{ref}}{\sigma_p}$$

Eq. 3

$$\zeta = \frac{x_{lab} - X_{ref}}{\sqrt{u_{ref}^2 + u_{lab}^2}}$$

Eq. 4

Where: x_{lab} is the measurement result reported by a participant

X_{ref} is the reference value (assigned value)

u_{ref} is the standard uncertainty of the reference value

u_{lab} is the standard uncertainty reported by a participant

σ_p is the standard deviation for proficiency assessment

The interpretation of the *z-* and *ζ-score* is done as follows (according to ISO/IEC 17043 [6]):

Satisfactory performance, |score| ≤ 2

Questionable performance,   2 < |score| < 3

Unsatisfactory performance, |score| ≥ 3

The z-score compares the participant's deviation from the reference value with the standard deviation for proficiency assessment (σ_p) used as common quality criterion. σ_p is defined by the PT organiser as the maximum acceptable standard uncertainty.

The ζ-score states if the laboratory result agrees with the assigned value within the respective uncertainty. The denominator is the combined uncertainty of the assigned value and the measurement uncertainty as stated by the laboratory. The ζ-score is therefore the most relevant evaluation parameter, as it includes all parts of a measurement result, namely the expected value (assigned value), its uncertainty and the unit of the result as well as the uncertainty of the reported values. An unsatisfactory ζ-score can either be caused by an inappropriate estimation of the concentration or of its uncertainty or both.

The standard uncertainty of the laboratory (u_{lab}) was estimated by dividing the reported expanded uncertainty by the reported coverage factor, *k*. When no uncertainty was reported, it was set to zero ($u_{lab} = 0$). When *k* was not specified, the reported expanded uncertainty was considered as the half-width of a rectangular distribution; u_{lab} was then calculated by dividing this half-width by √3, as recommended by Eurachem and CITAC [14].

Uncertainty estimation is not trivial; therefore an additional assessment was provided to each laboratory reporting uncertainty, indicating how reasonable their uncertainty estimate is. The standard uncertainty from the laboratory (u_{lab}) is most likely to fall in a range between a minimum uncertainty (u_{min}), and a maximum allowed (u_{max}, case "*a*"). u_{min} is set to the standard uncertainty of the reference value (u_{ref}). It is unlikely that a laboratory carrying out the analysis on a routine basis would measure the measurand with a smaller uncertainty than the expert laboratories chosen to establish the assigned value. u_{max} is set to the standard deviation (σ_p) accepted for the PT assessment.

If u_{lab} is smaller than u_{min} (case "*b*") the laboratory may have underestimated its uncertainty. However, such a statement has to be taken with care as each laboratory reported only measurement uncertainty, whereas the uncertainty of the reference value also includes contributions of homogeneity and stability. If those are large, measurement uncertainties smaller than u_{min} (u_{ref}) are possible and plausible.

If u_{lab} is larger than u_{max}, (case "*c*") the laboratory may have overestimated the uncertainty. An evaluation of this statement can be made when looking at the difference of the reported value and the assigned value: if the difference is small and the uncertainty is large, then overestimation is likely. If, however, the deviation is large but is covered by the uncertainty, then the uncertainty is properly assessed, but large. It should be pointed out that u_{max} is only a normative criterion if set down by legislation.

6.2   General observations

Results were received from 37 of the 38 registered laboratories. Those reporting "less than X" values were not evaluated. However, reported "less than X" values were compared with the corresponding $X_{ref} - U_{ref}$. If the reported limit value "X" was lower than the corresponding $X_{ref} - U_{ref}$, this statement should be considered incorrect, since the laboratory should have detected the respective element. In all laboratories in which "lower than X" was reported, X equals the reported limit of detection (LoD).

6.3   Laboratory results and scorings

Annexes 7-11 present the reported results as a table and as a graph. Furthermore, it includes the corresponding Kernel density plot, obtained using the software available from the Statistical Subcommittee of the Analytical Methods Committee of the UK Royal Society of Chemistry [15].

Figure 1 presents an overview of the *z*- and *ζ*-scores. From 81 (iAs) to 97 % (Cd) of the laboratories performed satisfactorily (*z*-score ≤ 2) for all the measurands investigated. Similarly, 69 (As) to 84 % (Cd) of the laboratories obtained satisfactory *ζ*-scores.

The assessment of reported uncertainties presented in Table 3 is based on the three uncertainty categories defined in chapter 6.1: "*a*" (realistic), "*b*" (underestimated) and "*c*" (overestimated). Most of the laboratories having reported underestimated uncertainties obtained unsatisfactory *ζ*-scores (Annexes 7-11).

Figure 1*: Overview of scores (in % and in the number of laboratories having satisfactory, questionable and unsatisfactory performance)*

Table 3 *– Uncertainty assessment (in %).*

	Case "a"	Case "b"	Case "c"
Total As	69	9	22
Total Cd	54	16	30
Total Hg	58	12	30
Total Pb	67	18	15
iAs	63	6	31

It is worth mentioning that most of the participants estimated correctly their measurement uncertainty for total As, total Pb and for inorganic arsenic. However, a relatively high percentage of participants overestimated their measurement uncertainty (particularly for the total Cd, total Hg and iAs). Two of the laboratories which overestimated their uncertainty (L07 and L26) were identified having reported uncertainties in percentages instead of mg kg^{-1}.

Particularly interesting are the results reported for iAs. In previous years the EURL-HM organized several PTs, upon request by DG SANCO, to check the analytical capabilities of laboratories to analyse iAs in different matrices: IMEP-107 (rice), IMEP-109 (seafood) and IMEP-112 (wheat, spinach and algae). During the initial screening, the University of Barcelona observed relatively high total As mass fraction in the test item used in IMEP-116. Therefore the EURL-HM decided to include iAs as an additional measurand. As the intake of mushrooms is high among some European populations it was considered interesting to check which percentage of total As was present as the toxic form iAs and whether NRLs were capable of providing satisfactory results for that measurand. The first thing that needs to be highlighted is the relatively high number of NRLs (sixteen) having reported results for this measurand. The second relevant information is the high percentage of satisfactory results reported taking into consideration the challenge of speciation analyses compared to the analysis of the total mass fraction of an element.

6.4   Further information extracted from the questionnaire

In addition to the submission of results, participants were asked to answer a number of questions related to:

> i) The analytical method used,
>
> ii) The quality assurance of their results

In order to allow the identification of all major potential sources of variability among the reported results we investigated the relation between each reported value (for each measurand) and the set of responses provided in the questionnaire. The statistical data treatment was performed using The Unscrambler X 10.1 (CAMO Software AS, Norway). Answers were first transformed into numerical variables, before applying partial least square regression modelling (*PLS-R*). Multivariate models succeed to "explain" a reasonable percentage of the total covariance relating the reported results and the set of answers. Furthermore, the model errors were generally lower than the observed variability for each corresponding set of reported values (expressed as the respective standard deviation). Therefore the multivariate models allowed reliable interpretations. As can be observed from Annexes 7-11, no significant differences have been observed among the participants. Nevertheless, as a general observation, higher reported results seem to be correlated to the following parameters:

  i)   Use microwave with HNO_3 and H_2O_2 for sample digestion,
  ii) Have experience with this type of analyses (analyse regularly identical matrix and analyte),
  iii) Quality assurance issues (e.g. having a quality system, being accredited, use CRMs for validation and/or instrument calibration and take part regularly in appropriate interlaboratory comparisons).

Furthermore, the technique used was also scrutinized. For the determination of the total As mass fraction most of the participants using atomic absorption spectrometry based techniques (particularly when combined with hydride generation techniques and microwave with HNO_3 and H_2O_2 for sample digestion) reported values slightly lower than those reported by participants using ICP-MS, maybe due to the presence of some organic species of As difficult to mineralize (e.g. arsenobetain). The high temperatures reached in the plasma would eliminate that problem when ICP-MS is used. No significant difference between the different techniques could be observed for the other measurands.

All participants except one, stated to have corrected their results for the water content. The residual moisture content of the test material was determined at IRMM following the protocol provided to the participants. The moisture content found was 3.94 % (value which agrees with the value obtained by Karl Fisher determination (3.95 %)). The average of all the reported values by the participants was 3.68 % (± 1.35 %, one standard deviation). Thus, participants have, in general, estimated correctly the moisture content and no significant influence could be observed while reporting their measurements related to dry mass.

Annex 12 summarizes all answers related with the experimental details and scorings (Q2.1, Q08, Q09, Q11, Q12.1 and Q16 in the questionnaire).

Table 4 presents the feedback received from the participants. Most of them are related to the amount of test material dispatched by the PT organiser and available for analysis.

***Table 4** – Feedback from participants (as taken from the questionnaire)*

Lab Code	Do you have any comments? Please let us know…
L01	The sample weight was very small
L02	consensus value for iAs in 1586a used
L05	The iAS analysis was unsuccesful and we have no material to repeat it.
L06	Sample amount was small.
L17	none
L19	A larger amount of test material would be desirable.
L22	The Cadmium results were at the upper end of the Cd calibration range.
L28	Question 12.1: d) 250-1000 samples/year (i.o. > 1000)
L34	The LOD for the As, Hg are not experimentally determined yet because the method is under validation by ICP-MS. We used internal std only for As, Hg with ICP-MS. We noticed that the amount of the PT usually is not enough.

7   Conclusion

The performance of NRLs for the determination of all investigated trace elements in freeze dried mushrooms should be considered satisfactory with z-scores ranging from 81 to 97 %. Thus, the analytical capability of NRLs for the determination of the investigated food contaminants, at the investigated levels of concentration, was successfully demonstrated. Indeed, when comparing the present performance with the one obtained in IMEP-39 (parallel PT, open to all food control laboratories on the same proficiency test item and applying the same performance criteria) the overall rates of satisfactory performance, obtained by the NRLs (expressed as z-scores) ranged 10 % to 25 % higher than the same rates in IMEP-39.

Quite a high percentage of satisfactory results were reported for iAs indicating that along the years the network of NRLs has improved its analytical capabilities in this field.

Participants using microwave-assisted digestion with nitric acid and hydrogen peroxide reported, in general, higher values than the others. This indicates that this method enables a more efficient sample digestion for the investigated food contaminants.

The technique used seems to have influenced the results for the determination of the total As whereby the majority of the participants which used AAS-based techniques reported lower values than participants using ICP-MS. This could be due to the high temperatures reached in the plasma.

8   Acknowledgements

The laboratories participating in this exercise, listed below, are kindly acknowledged. P. Conneely (IRMM) is acknowledged for the measurements performed to estimate the water content of the test samples. M-F. Tumba-Tshilumba is acknowledged for the particle size analysis measurements. F. Ulberth (IRMM) is acknowledged for reviewing the manuscript.

Organisation	Country
AGES	AUSTRIA
CODA-CERVA	BELGIUM
Central Laboratory for Chemical Testing and Control	BULGARIA
State General Laboratory	CYPRUS
ÚKZÚZ	CZECH REPUBLIC
State Veterinary Institute Olomouc	CZECH REPUBLIC
National Food Institute DTU	DENMARK
Danish Veterinary and Food Administration	DENMARK
Agricultural Research Centre	ESTONIA
Estonian Veterinary and Food Laboratory	ESTONIA
Finnish Customs Laboratory	FINLAND
Finnish Food Safety Authority Evira	FINLAND
ANSES LSA CIME	FRANCE
Laboratoire SCL de Bordeaux - FRANCE	FRANCE
Federal Office of Consumer Protection and Food Safety (BVL)	GERMANY
Regional Center of Plant Protection and Quality Control of Magnesia	GREECE
General Chemical State Laboratory	GREECE
National Food Chain Safety Office, Food and Feed Safety Directorate	HUNGARY
Health Service Executive	IRELAND
Istituto Zooprofilattico Sperimentale - TURIN	ITALY
Istituto Superiore di Sanità	ITALY
Institute of Food Safety, Animal Health and Environment	LATVIA
National Food and Veterinary Risk Assessment Institute	LITHUANIA
Scientific Institute of Public Health	LUXEMBURG
Environmental Health Directorate	MALTA
RIKILT	NETHERLANDS
Food Safety Autoryty of the Netherlands	NETHERLANDS
NIFES	NORWAY
National Institute of Public Health	POLAND
State Veterinary and Food Institute	SLOVAKIA
Zavod za Zdravstveno Varstvo MARIBOR	SLOVENIA
National Veterinary Institute	SLOVENIA
Laboratorio Arbitral Agroalimentario	SPAIN
National Food Agency	SWEDEN
The Food and Environment Research Agency	UNITED KINGDOM

9   Abbreviations

AAS	Atomic absorption spectroscopy
CITAC	Cooperation on international traceability in analytical chemistry
CV-AFS	Cold-vapour atomic fluorescence spectrometry
EU	European Union
ET-AAS	Electrothermal atomic absorption spectrometry
EURL-HM	European Union Reference Laboratory for Heavy Metals in Feed and Food
FI-HGAAS	Flow injection hydride-generation atomic aborption spectrometry
GUM	Guide to the expression of Uncertainty in Measurement
HPLC-ICPMS	High performance liquid chromatography inductively-coupled plasma mass spectrometry
IRMM	Institute for Reference Materials and Measurements
ILC	Interlaboratory Comparison
IMEP	International Measurement Evaluation Programme
ISO	International Organisation for Standardisation
ICP-MS	Inductively-coupled plasma mass spectrometry
ICP-SF-MS	Inductively-coupled plasma sector field mass spectrometry
JRC	Joint Research Centre
NRL	National Reference Laboratory
PT	Proficiency testing
PLS-R	Partial least squares regression
SS-CVAAS	Solid sampling cold-vapour atomic aborption spectrometry

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Annexes

Annex 1: Invitation letter to NRLs

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements
European Union Reference Laboratory for Heavy Metals

Geel, 9 September 2013
JRC.D.5/PRO/FCR/acs/ARES

Subject : Proficiency Test for EURL Heavy Metals in Feed and Food

Dear National Reference Laboratory representative,

We would like to invite you on behalf of the EURL Heavy Metals in Feed and Food, to participate in the Proficiency Test IMEP-116 for the **"Determination of total Cd, Pb, As, Hg and inorganic As in mushrooms"**.

You are kindly reminded that according to Regulation (EC) No 882/2004 it is your duty as NRL to participate in PTs organised by the EURL-HM if you hold a mandate for the type of matrix investigated.

Your participation is free of charge.

Please register electronically for this proficiency test round using the following link:

https://web.jrc.ec.europa.eu/ilcRegistrationWeb/registration/registration.do?selComparison=1040

Once you have submitted your registration electronically, please (1) print your registration form, (2) sign it, and (3) fax it to us. Your fax is the confirmation of your participation.

The **deadline for registration is 30 April 2013**. Samples will be sent to participants during the first half of May 2013. The deadline for submission of results is **15 June 2013**.

Do not hesitate to contact us, in case of questions/doubts,

Yours sincerely

Dr. Fernando Cordeiro
IMEP-116 Coordinator

Dr. Piotr Robouch
Operating Manager EURL-HM

Cc: Franz Ulberth (Head of Unit SFB)

Retieseweg 111, B-2440 Geel - Belgium. Telephone: +32-(0)14-571 211.
Telephone: direct line +32-(0)14-571 687, Fax: +32-(0)14-571 865.

E-mail: jrc-irmm-crl-heavy-metals@ec.europa.eu
Web site: http://irmm.jrc.ec.europa.eu

Annex 2: IRMM – IMEP web announcement

The page content is a rotated (landscape) screenshot of a web page from the Joint Research Centre, Institute for Reference Materials and Measurements (IRMM).

EUROPA > European Commission > JRC > IRMM > EU Reference Laboratories > EURL heavy metals > Interlaboratory comparisons > IMEP-116

JOINT RESEARCH CENTRE
Institute for Reference Materials and Measurements (IRMM)

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IMEP-116: Total Cd, Pb, As, Hg and inorganic As in mushrooms

The IMEP-116 interlaboratory comparison (ILC) exercise focuses on the analysis of total cadmium, lead, arsenic, mercury and inorganic arsenic in mushrooms. This ILC is organised in the frame of EU Regulation 1881:2006, which sets maximum levels for certain contaminants in foodstuffs and contributes to the implementation of high quality and uniform analytical results.
The main objective of this exercise is to evaluate the capabilities of nominated National Reference Laboratories (NRLs) in the determination of heavy metals in mushrooms, which maximum limits are set according to EU Regulation 1881:2006. It is therefore a proficiency test (PT).
Participation in IMEP-116 is mandatory for all NRLs having experience in this kind of analysis.

Only appointed NRLs can participate in this exercise.

Registration is free of charge.

Please register using the following link: https://web.jrc.ec.europa.eu/ilcRegistrationWeb/registration/registration.do?selComparison=1040

Test materials and analytes

The test material to be analysed is freeze dried mushroom contained in a glass bottle. Each participant will receive one bottle of the test item.
The measurands are total Cd, Pb, As, Hg and inorganic As in mushrooms.

General outline of the exercise

Participants are requested to perform 1 - 3 independent analyses using the method of their choice, and to report their result for the PT assessment, its associated measurement uncertainty and coverage factor k.
Detailed instructions will be sent together with the test sample.

Schedule

Registration	Sample dispatch	Reporting of results	Report to participants
Deadline 30/04/2013	May 2013	Deadline 22/06/2013	November 2013

Latest update 3 June, 2013

News archive
- Environmental analysis
- Nuclear research
- Reference materials and measurements
- Food, biotechnology and health

JRC-IRMM video

You Tube

IRMM catalogue

TrainMiC

ERM

EURL

Annex 3: Sample accompanying letter

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements
European Union Reference Laboratory for Heavy Metals

Geel, 15 May 2013
JRC.D5/FCR/acs/

Participation in IMEP-116, a proficiency test exercise for the determination of total cadmium (Cd), lead (Pb), arsenic (As), mercury (Hg) and inorganic arsenic (iAs) in mushrooms

Dear XXXXXX,

Thank you for participating in the IMEP-116 proficiency test for the determination of the total Cd, Pb, Ar, Hg and iAs in mushrooms. This proficiency test (PT) exercise is organised in support to the EU Regulation 1881:2006 which sets maximum levels for certain contaminants in foodstuffs.

Please keep this letter. You need it to report your results.

This parcel contains:
a) One bottle containing approximately 2.5 g of the test material
b) A "Confirmation of Receipt" form
c) This accompanying letter.

Please check whether the bottle containing the test material remained undamaged during transport. Then, please send the "Confirmation of receipt" form back (fax: +32-14-571865, e-mail: jrc-irmm-imep@ec.europa.eu). You should store the sample in a dark place at ≤4 ºC until analysis.

The measurand is total Cd, Pb, Ar, Hg and iAs in mushroom. The sample matrix is a mixture of commercially available mushroom (freeze dried).

The procedure used for the analyses should resemble as closely as possible the one that you use in routine analyses.

The results are to be reported with correction for moisture (in dry mass).

To calculate the **water content** in the test material, please apply the following procedure:

Retieseweg 111, B-2440 Geel - Belgium. Telephone: +32-(0)14-571 211.
Telephone: direct line +32-(0)14-571 687, Fax: +32-(0)14-571 865.

E-mail: jrc-irmm-crl-heavy-metals@ec.europa.eu
Web site: http://irmm.jrc.ec.europa.eu

(1) Weigh approximately 500 mg of test material in a petri-dish of 3.5 cm diameter, preferably with a lid. The thickness of the powder-layer should be about 3-4 mm covering the bottom of the dish.

(2) Place it in a checked and calibrated drying oven at 90 ± 2 °C for 60 ± 2 minutes. Allow the glass container (covered with the lid) to cool down for about 30 minutes in a desiccator before weighing.

(3) Calculate the average mass loss from the dried material in percentage of the initial mass.

Please note that this drying method is devised to result in a mass loss that corresponds to the water content in % (m/m) as measured by Karl Fischer titration which is specific for water. Therefore it is not necessary to dry and continue weighing until constant mass. Keeping the material longer than one hour in the oven will result in an excessive mass loss and an erroneous dry-mass correction.

Note : do not use for the heavy metal determinations the aliquots of test material that you have used for the water content determination!

Reporting of results

Please perform two or three independent measurements, correct the measurements results for recovery and for the moisture content and report on the reporting website:

- the **mean** of your two or three measurement results (mg kg^{-1}, as dry mass)
- the associated expanded **uncertainty** (mg kg^{-1}),
- the **coverage factor** and
- the **technique** you used.

The results should be reported in the same form (e.g. number of significant figures) as those normally reported to the customer.

The reporting website is https://irmm.jrc.ec.europa.eu/ilc/ilcReporting.do

To access the webpage you need a personal password key, which is: xxxxxxxx. The system will guide you through the reporting procedure. After entering your results, please complete also the relating questionnaire.

Do not forget to submit and confirm always when required.

Directly after submitting your results and the questionnaire information online, you will be prompted to print the completed report form. Please do so, **sign the paper version and return it to IRMM by fax (at +32-14-571-865) or by e-mail**. Check your results carefully for any errors before submission, since this is your last definitive confirmation.

Retieseweg 111, B-2440 Geel - Belgium. Telephone: +32-(0)14-571 211.
Telephone: direct line +32-(0)14-571 687, Fax: +32-(0)14-571 865.

E-mail: jrc-irmm-crl-heavy-metals@ec.europa.eu
Web site: http://irmm.jrc.ec.europa.eu

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The **deadline** for submission of results is **22/06/2013**.

Please keep in mind that collusion is contrary to professional scientific conduct and serves only to nullify the benefits of proficiency tests to customers, accreditation bodies and analysts alike.

Your participation in this project is greatly appreciated. If you have any remaining questions, please contact me by e-mail: jrc-irmm-imep@ec.europa.eu

With kind regards

Fernando Cordeiro Pepper

Dr. Fernando Cordeiro
IMEP-116 Coordinator

Enclosures: 1) One bottle containing the test material; 2) Confirmation of receipt form; 3) Accompanying letter.

Cc: F. Ulberth

Annex 4: Confirmation of receipt form

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements
International Measurement Evaluation Program

Annex to
JRC.D5/FCR/acs/ARES(2013)

«Title» «Firstname» «Surname»
«Organisation»
«Department»
«Address»
«Address2»
«Zip» «Town»
«Country»

IMEP-116

Total cadmium (Cd), lead (Pb), arsenic (As), mercury (Hg) and inorganic arsenic (iAs) in mushrooms

Confirmation of receipt of the samples

Please return this form at your earliest convenience.
This confirms that the sample package arrived.
In case the package is damaged,
please state this on the form and contact us immediately.

ANY REMARKS ……………………………………………………………………………………………
……………………………………………………………………………………………

Date of package arrival ……………………………….

Signature ……………………………….

Please return this form to:

Fernando Cordeiro Raposo

IMEP-116 Coordinator
EC-JRC-IRMM
Retieseweg 111
B-2440 GEEL, Belgium

Fax   : +32-14-571865

e-mail : JRC-IRMM-CRL-HEAVY-METALS@ec.europa.eu

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211
Telephone: direct line (32-14) 571 687, Fax: (32-14) 571 865

E-mail: jrc-irmm-crl-heavy-metals@ec.europa.eu
Web site: http://irmm.jrc.ec.europa.eu

Annex 5: Questionnaire

1. **Did you correct your measurements for the analytical recovery?**

   ○ a) Yes
   ○ b) No

2. **How did you determine the analytical recovery?**

   ☐ a) adding a known amount of the same analyte (spiking)
   ☐ b) using a certified reference material
   ☐ c) other

   2.1. If other, please specify:

   [   ]

   2.1.1. Please fill the table:

   Analytical recovery (in %) and limit of detection, LoD (in mg/Kg)

Questions/Response table	Total Cd	Total Pb	Total As	Total Hg	Inorganic As
Recovery (in %)					
LoD (in mg/Kg)					

3. **What is the level of confidence (in %) reflected by the coverage factor k assigned to your measurement uncertainty?**

   [   ]

4. **What is the basis of your measurement uncertainty estimation (multiple answers are possible)?**

   ☐ a) Uncertainty budget (ISO-GUM)
   ☐ b) Known uncertainty of the standard method (ISO 21748)
   ☐ c) Uncertainty of the method (in-house validation)
   ☐ d) Measurement of replicates (precision)
   ☐ e) Estimation based on judgemnt
   ☐ f) From interlaboratory comparison data
   ☐ g) Other

   4.1. If other, please specify

   [   ]

5. **Do you usually provide an uncertainty statement to your customers for this type of analysis?**

   ○ a) Yes
   ○ b) No

6. **Did you correct for the moisture content of the sample?**

   ○ a) Yes
   ○ b) No

   6.1. If Yes, what is the moisture content (in % of the sample mass)?

   [   ]

   6.2. If no, what was the reason not to do this?

   [   ]

7. **Did you analyse the sample according to an official method?**

   ○ a) Yes
   ○ b) No

   7.1. If Yes, which one?

Questions/Response table	Which offcial method?
Total Cd	
Total Pb	
Total As	
Total Hg	
Inorganic As	

8. **Did you use microwave digestion techniques?**

   ○ a) Yes
   ○ b) No

   8.1. Any comment regarding this question?

   [   ]

9. **Did you use HNO3 and H2O2 for sample digestion?**

  ○ a) Yes
  ○ b) No

9.1. Any comment regarding this question?

[   ]

10. **Did you use an internal standard?**

  ○ a) Yes
  ○ b) No

11. **Other experimental details:**

Please provide a short description for each experimental issue.

Questions/Response table	Sample pre-treatment	Digestion	Extraction/separation	Instrumental calibration
Total Cd				
Total Pb				
Total As				
Total Hg				
Inorganic As				

12. **Does your laboratory carry out this type of analysis (as regards the analytes, matrix and methods) on a regular basis?**

  ○ a) Yes
  ○ b) No

12.1. If Yes, please fill the corresponding table cell (samples/year)

Questions/Response table	b) 0-50	c) 50-250	d) > 1000	e) more than 1000	Info
Total Cd	☐	☐	☐	☐	
Total Pb	☐	☐	☐	☐	
Total Hg	☐	☐	☐	☐	
Total As	☐	☐	☐	☐	
Inorganic As	☐	☐	☐	☐	

13. **Does your laboratory have a quality system in place?**

  ○ a) Yes
  ○ b) No

13.1. If Yes, which:

  ☐ a) ISO 17025
  ☐ b) ISO 9000 series
  ☐ c) Other

  13.1.1. If other, please specify

  [   ]

14. **Is your laboratory accredited for this type of analysis?**

Questions/Response table	Accredited	Not accredited	Info
Total Cd	☐	☐	
Total Pb	☐	☐	
Total As	☐	☐	
Total Hg	☐	☐	
Inorganic As	☐	☐	

15. Does your laboratory take part in interlaboratory comparison scheme for this type of analysis?

   ○ a) Yes
   ○ b) No

15.1. If yes, which one(s)

     Interlaboratory comparison (ILC) for:

Questions/Response table	ILC (please identify it)
Total Cd	
Total Pb	
Total As	
Total Hg	
Inorganic As	

16. Does your laboratory use a certified reference material (CRM) for this type of analysis?

    CRM used and purpose

Questions/Response table	CRM used	Validation of procedures	Instrument calibration
Total Cd			
Total Pb			
Total As			
Total Hg			
Inorganic As			

17. Do you have any comments? Please let us know: ...

Annex 6: Homogeneity and stability studies

6.1 Homogeneity studies

Homogeneity	*Total As*		*Total Cd*		*Total Hg*		*Total Pb*	
Bottle ID	*R₁*	*R₂*	*R₁*	*R₂*	*R₁*	*R₂*	*R₁*	*R₂*
3	0.539	0.558	3.88	3.95	0.0798	0.0808	0.246	0.244
37	0.531	0.524	3.83	3.95	0.0846	0.0846	0.234	0.296
52	0.568	0.532	3.99	3.89	0.0846	0.0842	0.232	0.236
60	0.558	0.536	3.84	3.94	0.0842	0.0835	0.237	0.244
97	0.523	0.555	3.88	3.91	0.0842	0.0819	0.242	0.260
113	0.528	0.562	4.07	3.92	0.0852	0.0822	0.250	0.259
138	0.535	0.554	3.87	4.06	0.0832	0.0877	0.241	0.258
141	0.558	0.552	3.93	3.98	0.0819	0.0828	0.239	0.247
174	0.554	0.548	3.91	3.96	0.0863	0.0817	0.237	0.238
194	0.554	0.562	3.96	3.84	0.0783	0.0785	0.235	0.292
Homogeneity assessment according to ISO 13528 [7]								
Mean	0.546		3.93		0.0830		0.248	
σ_p	0.082		0.393		0.0125		0.050	
0.3* σ_p	0.025		0.118		0.0037		0.015	
s_x	0.009		0.036		0.0021		0.010	
s_w	0.016		0.077		0.0017		0.020	
s_s	0.000		0.000		0.0018		0.000	
$s_s \leq 0.3 * \sigma_p$	**Pass**		**Pass**		**Pass**		**Pass**	

Where σ_p is the standard deviation for the PT assessment,

s_x is the standard deviation of the sample averages,

s_w is the within-sample standard deviation,

s_s is the between-sample standard deviation,

6.2 Stability studies

		Time in Weeks					
	0	**3**	**5**	**8**	Slope of linear regression significantly <> 0 (95%) : No		
As	0.576	0.57	0.571	0.547	Standard error of the slope =	0.002	
	0.542	0.534	0.565	0.564	**Uncertainty contribution**	u_{st} =	**0.015**
					Slope of linear regression significantly <> 0 (95%) : No		
Cd	3.94	4.03	3.86	3.99	Standard error of the slope =	0.008	
	3.92	3.92	4.03	3.9	**Uncertainty contribution**	u_{st} =	**0.060**
					Slope of linear regression significantly <> 0 (95%) : No		
Hg	0.0849	0.0814	0.0833	0.0861	Standard error of the slope =	0.000	
	0.0807	0.0833	0.0845	0.0839	**Uncertainty contribution**	u_{st} =	**0.002**
					Slope of linear regression significantly <> 0 (95%) : No		
Pb	0.243	0.267	0.262	0.244	Standard error of the slope =	0.001	
	0.242	0.262	0.252	0.245	**Uncertainty contribution**	u_{st} =	**0.010**

Annex 7: Results for total As

Assigned range: X_{ref} = 0.646; U_{ref} = 0.048 (k=2); σ_p = 0.10 (all values in mg kg^{-1})

Lab Code	X_{lab}	U_{lab}	k^a	Technique	u_{lab}	z-score[b]	ζ-score[b]	Unc.[c]
L02	0.61	0.07	2	ICP-MS	0.035	-0.4	-0.8	a
L03	0.496	0.05	2	HG-AAS	0.025	-1.5	-4.3	a
L05	0.386	0.097	2	HG-AAS	0.0485	-2.7	-4.8	a
L06	0.636	0.197	2	ICP-MS	0.0985	-0.1	-0.1	c
L07	0.56	6	2	ICP-MS	3	-0.9	0.0	c
L08	0.51	0.1	2	ICP-MS	0.05	-1.4	-2.5	a
L10	0.57	0.11	2	ICP-MS	0.055	-0.8	-1.3	a
L11	0.557	0.111	2	ICP-MS	0.0555	-0.9	-1.5	a
L12	< 0.85			ETAAS				
L13	0.434	0.184	2	HG-AAS	0.092	-2.2	-2.2	a
L14	0.646	0.065	2	ICP-MS	0.0325	0.0	0.0	a
L15	0.598	0.12	2	ICP-MS	0.06	-0.5	-0.7	a
L16	0.621	0.076	2	HG-AAS FIAS	0.038	-0.3	-0.6	a
L17	0.543	0.058	2	ICP-MS	0.029	-1.1	-2.7	a
L18	0.577	0.098	2	ICP-MS	0.049	-0.7	-1.3	a
L20	0.533	0.008	2	ICP-MS	0.004	-1.2	-4.7	b
L21	0.61	0.11	2	ICP-MS	0.055	-0.4	-0.6	a
L22	0.59	0.29	2	ICP-MS	0.145	-0.6	-0.4	c
L23	0.62	0.099	2	ICP-MS	0.0495	-0.3	-0.5	a
L24	0.77	0.31	2	ICP/AES	0.155	1.3	0.8	c
L25	0.82	0.25	2	ETAAS	0.125	1.8	1.4	c
L26	0.56	8	2	ICP-MS	4	-0.9	0.0	c
L27	0.63	0.252	2	ICP-MS	0.126	-0.2	-0.1	c
L28	0.63	0.15	2	ICP-MS	0.075	-0.2	-0.2	a
L29	0.61	0.11	2	ICP-MS	0.055	-0.4	-0.6	a
L30	0.5	0.1	2	ETAAS	0.05	-1.5	-2.6	a
L31	0.503	0.101	√3	ICP-MS	0.058312	-1.5	-2.3	a
L32	0.611	0.056	2	ICP-MS	0.028	-0.4	-0.9	a
L33	0.625	0.072	2	AAS	0.036	-0.2	-0.5	a
L34	0.58	0.092	2	ICP-MS	0.046	-0.7	-1.3	a
L35	0.22	0.013	2	HG-AAS	0.0065	-4.4	-17.2	b
L36	0.725			ICP-MS	0	0.8	3.3	b
L37	0.575	0.081	2	ICP-MS	0.0405	-0.7	-1.5	a

[a] √3 is set by the ILC coordinator when no expansion factor *k* is reported. The reported uncertainty was assumed to have a rectangular distribution with $k = \sqrt{3}$.

[b] **Satisfactory, Questionable, Unsatisfactory**

[c] **a** : u_{min} (u_{ref}) ≤ u_{lab} ≤ u_{max} (σ_p);   **b** : u_{lab} < u_{min};   and **c** : u_{lab} > u_{max} (σ_p)

IMEP-116: Total arsenic in mushroom

$X_{ref} = 0.646$; $U_{ref} = 0.048$ ($k=2$); $\sigma_p = 0.10$ (mg kg^{-1})
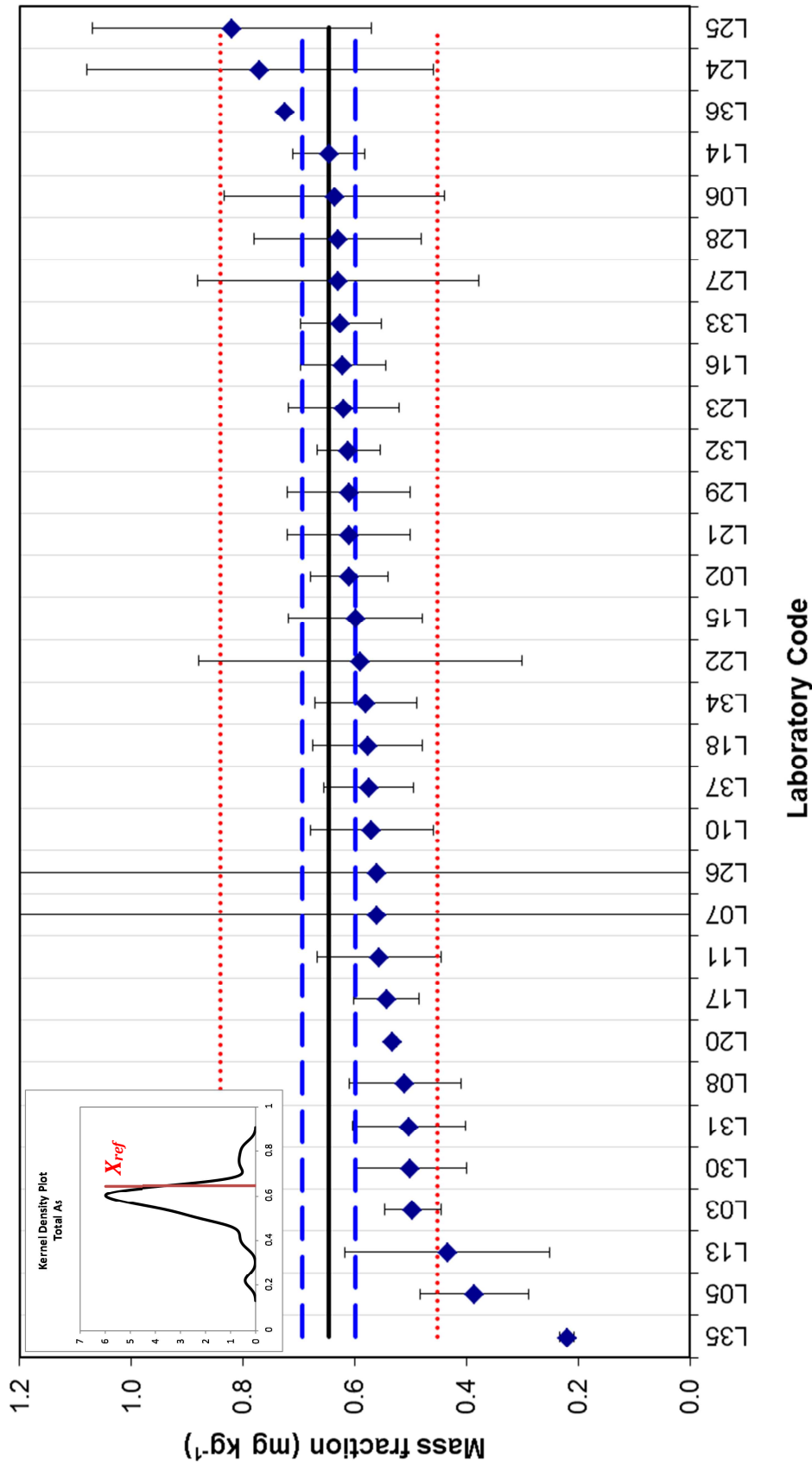
Laboratory Code

Mass fraction (mg kg^{-1})

Measurement results and associated uncertainties (reported uncertainties shown).

Reference value (X_{ref}): solid black line; Reference interval ($X_{ref} \pm U_{ref}$): dashed blue lines; Target interval ($X_{ref} \pm 2\sigma_p$): dotted red lines.

Kernel Density Plot
Total As

X_{ref}

Annex 8: Results for total Cd

Assigned range: X_{ref} = 4.21; U_{ref} = 0.33 (k=2); σ_p = 0.42 (all values in mg kg^{-1})

| Lab Code	X_{lab}	U_{lab}	k [a]	Technique	u_{lab}	z-score [b]	ζ-score [b]	Unc. [c]
L01	3.68	0.81	2	ETAAS	0.405	-1.2	-1.2	a
L02	4	0.4	2	ICP-MS	0.2	-0.5	-0.8	a
L03	3.57	0.46	2	ETAAS	0.23	-1.5	-2.2	a
L04	4.01	0.38	2	AAS	0.19	-0.5	-0.8	a
L05	4.67	1	2	GF-AAS	0.5	1.1	0.9	c
L06	4.74	1.95	2	ICP-MS	0.975	1.3	0.5	c
L07	4	7	2	ICP-MS	3.5	-0.5	-0.1	c
L08	3.75	0.97	2	ICP-MS	0.485	-1.1	-0.9	c
L09	3.633	0.372	2	ETAAS	0.186	-1.4	-2.3	a
L10	4	0.96	2	ICP-MS	0.48	-0.5	-0.4	c
L11	3.72	0.67	2	ICP-MS	0.335	-1.2	-1.3	a
L12	4.5	0.72	2	ETAAS	0.36	0.7	0.7	a
L13	4.056	0.444	2	ETAAS	0.222	-0.4	-0.5	a
L14	3.85	0.39	2	ICP-MS	0.195	-0.8	-1.4	a
L15	4.19	0.84	2	ICP-MS	0.42	0.0	0.0	a
L16	3.84	0.34	2	ETAAS	0.17	-0.9	-1.5	a
L17	3.42	0.36	2	ICP-MS	0.18	-1.9	-3.2	a
L18	4.14	0.75	2	ICP-MS	0.375	-0.2	-0.2	a
L19	3.5	0.5	2	ICP-MS	0.25	-1.7	-2.4	a
L20	4.092	0.001	2	ICP-MS	0.0005	-0.3	-0.7	b
L21	4.24	1.08	2	ETAAS	0.54	0.1	0.1	c
L22	4.74	1.3	2	ICP-MS	0.65	1.3	0.8	c
L23	3.8	0.38	2	ICP-MS	0.19	-1.0	-1.6	a
L24	0.79	0.28	2	ETAAS	0.14	-8.1	-15.8	b
L25	3.848	0.885	2	ETAAS	0.4425	-0.8	-0.8	c
L26	3.52	5	2	ICP-MS	2.5	-1.6	-0.3	c
L27	4.2	1.68	2	ICP-MS	0.84	0.0	0.0	c
L28	4	0.67	2	ICP-MS	0.335	-0.5	-0.5	a
L29	3.81	0.49	2	ICP-MS	0.245	-0.9	-1.3	a
L30	3.85	0.8	2	ETAAS	0.4	-0.8	-0.8	a
L31	3.626	0.725	√3	ICP-MS	0.418579	-1.4	-1.3	a
L32	3.591	0.155	2	ICP-MS	0.0775	-1.5	-3.4	b
L33	3.915	0.129	2	AAS	0.0645	-0.7	-1.6	b
L34	3.9	0.64	2	AAS	0.32	-0.7	-0.8	a
L35	4.193	0.12	2	ETAAS	0.06	0.0	-0.1	b
L36	3.992			ICP-MS	0	-0.5	-1.3	b
L37	4.153	10	2	ICP-MS	5	-0.1	0.0	c

[a] √3 is set by the ILC coordinator when no expansion factor k is reported. The reported uncertainty was assumed to have a rectangular distribution with $k = \sqrt{3}$.

[b] **Satisfactory**, **Questionable**, **Unsatisfactory**

[c] **a** : u_{min} (u_{ref}) ≤ u_{lab} ≤ u_{max} (σ_p);   **b** : $u_{lab} < u_{min}$;   and **c** : $u_{lab} > u_{max}$ (σ_p)

IMEP-116: Total cadmium in mushroom

X_{ref} = 4.21 ; U_{ref} = 0.33 (k=2); σ_p = 0.42 (mg kg^{-1})
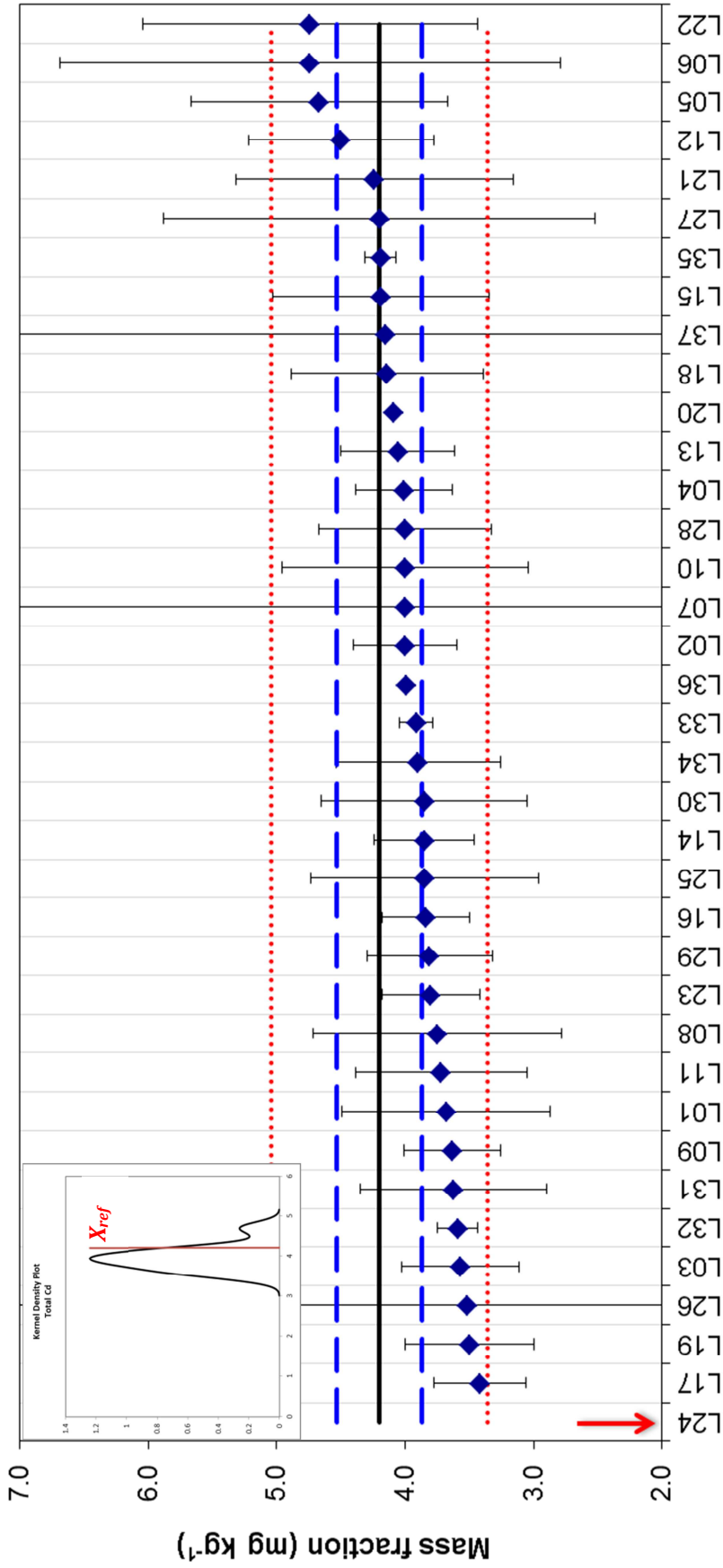
Measurement results and associated uncertainties (reported uncertainties shown).
Reference value (X_{ref}): solid black line; Reference interval ($X_{ref} \pm U_{ref}$): dashed blue lines; Target interval ($X_{ref} \pm 2\sigma_p$): dotted red lines.

Mass fraction (mg kg^{-1})

Laboratory Code

Annex 9: Results for total Pb

Assigned range: X_{ref} = 0.267; U_{ref} = 0.031 (k=2); σ_p = 0.05 (all values in mg kg^{-1})

Lab Code	X_{lab}	U_{lab}	k^a	Technique	u_{lab}	z-score[b]	ζ-score[b]	Unc.[c]
L01	0.168	0.042	2	ETAAS	0.021	-1.9	-3.8	a
L03	0.34	0.07	2	ETAAS	0.035	1.4	1.9	a
L04	0.13	0.027	2	AAS	0.0135	-2.6	-6.7	b
L05	0.1	0.035	2	GF-AAS	0.0175	-3.1	-7.1	a
L06	0.272	0.136	2	ICP-MS	0.068	0.1	0.1	c
L07	0.25	9	2	ICP-MS	4.5	-0.3	0.0	c
L08	0.207	0.058	2	ICP-MS	0.029	-1.1	-1.8	a
L09	0.292	0.071	2	ETAAS	0.0355	0.5	0.6	a
L10	0.23	0.064	2	ICP-MS	0.032	-0.7	-1.0	a
L11	0.211	0.042	2	ICP-MS	0.021	-1.0	-2.1	a
L12	< 1.8			ETAAS				
L13	0.146	0.064	2	ETAAS	0.032	-2.3	-3.4	a
L14	< 0.3			ICP-MS				
L15	0.22	0.044	2	ICP-MS	0.022	-0.9	-1.7	a
L16	0.252	0.042	2	ETAAS	0.021	-0.3	-0.6	a
L17	0.307	0.033	2	ICP-MS	0.0165	0.7	1.8	a
L18	0.242	0.032	2	ICP-MS	0.016	-0.5	-1.1	a
L19	0.31	0.1	2	ICP-MS	0.05	0.8	0.8	a
L20	0.12	0.005	2	ICP-MS	0.0025	-2.8	-9.3	b
L21	0.214	0.075	2	ETAAS	0.0375	-1.0	-1.3	a
L22	0.37	0.12	2	ICP-MS	0.06	1.9	1.7	c
L23	0.21	0.021	2	ICP-MS	0.0105	-1.1	-3.0	b
L24	0.27	0.1	2	ETAAS	0.05	0.1	0.1	a
L25	0.28	0.08	2	ETAAS	0.04	0.2	0.3	a
L26	0.234	6	2	ICP-MS	3	-0.6	0.0	c
L27	0.25	0.125	2	ICP-MS	0.0625	-0.3	-0.3	c
L28	0.22	0.04	2	ICP-MS	0.02	-0.9	-1.9	a
L29	0.282	0.074	2	ICP-MS	0.037	0.3	0.4	a
L30	0.2	0.04	2	ETAAS	0.02	-1.3	-2.6	a
L31	0.258	0.052	√3	ICP-MS	0.030022	-0.2	-0.3	a
L32	0.256	0.018	2	ICP-MS	0.009	-0.2	-0.6	b
L33	< 0.5			AAS				
L34	0.29	0.054	2	AAS	0.027	0.4	0.7	a
L35	0.285	0.012	2	ETAAS	0.006	0.3	1.1	b
L36	0.226			ICP-MS		-0.8	-2.6	b
L37	0.268	0.051	2	ICP-MS	0.0255	0.0	0.0	a

[a] √3 is set by the ILC coordinator when no expansion factor k is reported. The reported uncertainty was assumed to have a rectangular distribution with $k = √3$.

[b] **Satisfactory**, **Questionable**, **Unsatisfactory**

[c] **a** : u_{min} (u_{ref}) ≤ u_{lab} ≤ u_{max} (σ_p);   **b** : u_{lab} < u_{min};   and **c** : u_{lab} > u_{max} (σ_p)

IMEP-116: Total lead in mushroom

$X_{ref} = 0.267$; $U_{ref} = 0.031$ ($k=2$); $\sigma_p = 0.05$ ($mg\ kg^{-1}$)

Measurement results and associated uncertainties (reported uncertainties shown).
Reference value (X_{ref}): solid black line; Reference interval ($X_{ref} \pm U_{ref}$): dashed blue lines; Target interval ($X_{ref} \pm 2\sigma_p$): dotted red lines.

Annex 10: Results for total Hg

Assigned range: X_{ref} = 0.076; U_{ref} = 0.007 (k=2); σ_p = 0.011 (all values in mg kg^{-1})

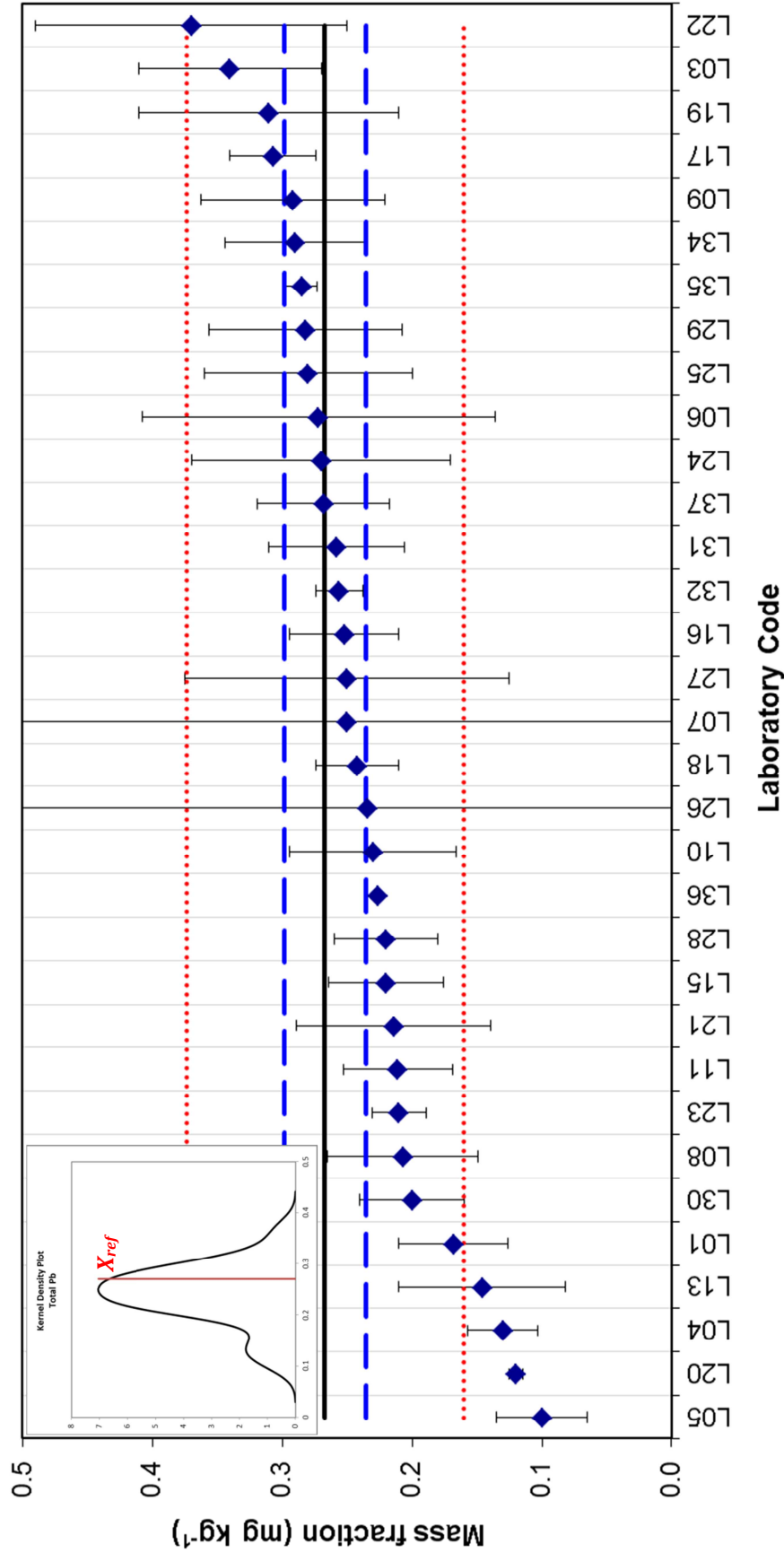| Lab Code	X_{lab}	U_{lab}	k^a	Technique	u_{lab}	z-score[b]	ζ-score[b]	Unc.[c]
L02	0.085	0.009	2	ICP-MS	0.0045	0.8	1.6	a
L03	0.0801	0.009	2	DMA	0.0045	0.4	0.7	a
L05	0.63	0.158	2	CV-AAS	0.079	48.5	7.0	c
L06	0.0962	0.0385	2	ICP-MS	0.01925	1.8	1.0	c
L07	0.089	10	2	ICP-MS	5	1.1	0.0	c
L08	0.07	0.020	2	ICP-MS	0.01	-0.5	-0.6	a
L10	0.061	0.012	2	DMA	0.006	-1.3	-2.2	a
L11	0.0843	0.0152	2	DMA	0.0076	0.7	1.0	a
L12	0.099	0.009	2	DMA	0.0045	2.0	4.0	a
L13	0.088	0.04	2	CV-AAS	0.0175	1.0	0.7	c
L14	0.083	0.012	2	DMA	0.006	0.6	1.0	a
L15	0.0739	0.015	2	CV-AAS	0.0075	-0.2	-0.3	a
L16	0.09	0.014	2	CV-AAS	0.007	1.3	1.9	a
L17	0.063	0.009	2	ICP-MS	0.0045	-1.1	-2.3	a
L18	0.085	0.013	2	ICP-MS	0.0065	0.8	1.2	a
L19	0.1	0.01	2	CV-AAS	0.005	2.1	3.9	a
L20	0.082	0.052	2	CV-AAS	0.026	0.5	0.2	c
L21	0.083	0.037	2	CV-AAS	0.0185	0.6	0.4	c
L22	0.077	0.015	2	ICP-MS	0.0075	0.1	0.1	a
L23	0.075	0.015	2	DMA	0.0075	-0.1	-0.1	a
L24	0.13	0.05	2	DMA	0.025	4.7	2.1	c
L25	0.08	0.02	2	CV-AAS	0.01	0.3	0.4	a
L26	0.0774	10	2	DMA	5	0.1	0.0	c
L27	0.076	0.0304	2	ICP-MS	0.0152	0.0	0.0	c
L28	0.075	0.016	2	DMA	0.008	-0.1	-0.1	a
L29	< 0.1			ICP-MS				
L30	0.086	0.02	2	CV-AAS	0.01	0.9	0.9	a
L31	0.0724	0.0036	√3	DMA	0.002078	-0.3	-0.9	b
L32	0.078	0.015	2	ICP-MS	0.0075	0.2	0.2	a
L33	0.072	0.004	2	CV-AAS	0.002	-0.4	-1.0	b
L34	0.11	0.039	2	ICP-MS	0.0195	3.0	1.7	c
L35	0.116	0.005	2	CV-AAS	0.0025	3.5	9.2	b
L36	0.089			ICP-MS	0	1.1	3.6	b
L37	0.083	0.012	2	ICP-MS	0.006	0.6	1.0	a

[a] √3 is set by the ILC coordinator when no expansion factor k is reported. The reported uncertainty was assumed to have a rectangular distribution with k = √3.

[b] **Satisfactory**, **Questionable**, **Unsatisfactory**

[c] **a** : u_{min} (u_{ref}) ≤ u_{lab} ≤ u_{max} (σ_p);   **b** : u_{lab} < u_{min};   and **c** : u_{lab} > u_{max} (σ_p)

IMEP-116: Total mercury in mushroom

$X_{ref} = 0.076$; $U_{ref} = 0.007$ (k=2); $\sigma_p = 0.011$ (mg kg^{-1})
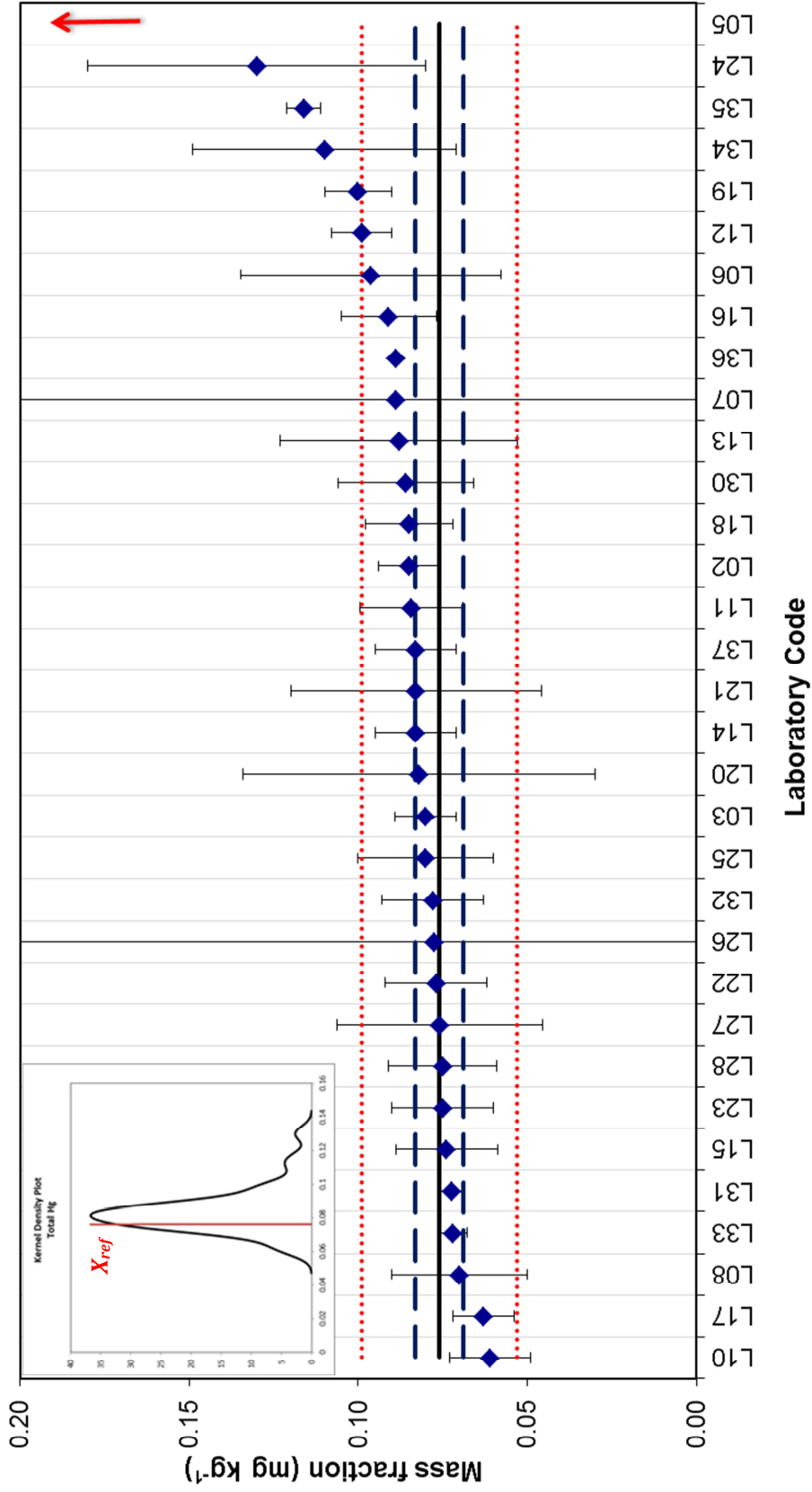
Mass fraction (mg kg^{-1})

Laboratory Code

Measurement results and associated uncertainties (reported uncertainties shown).
Reference value (X_{ref}): solid black line; Reference interval ($X_{ref} \pm U_{ref}$): dashed blue lines; Target interval ($X_{ref} \pm 2\sigma_p$): dotted red lines.

Annex 11: Results for inorganic As

Assigned range: X_{ref} = 0.321; U_{ref} = 0.026 (k=2); σ_p = 0.06 (all values in mg kg^{-1})

Lab Code	X_{lab}	U_{lab}	k^a	Technique	u_{lab}	z-score[b]	ζ-score[b]	Unc.[c]
L02	0.37	0.04	2	HPLC-ICP-MS	0.023094	0.8	1.8	a
L03	0.056	0.008	2	HG-AAS	0.004	-4.3	-19.8	b
L07	0.38	8	2	HPLC-ICP-MS	4	1.0	0.0	c
L08	0.42	0.084	2	ICP-MS	0.042	1.6	2.2	a
L10	0.51	0.13	2	LC-ICP-MS	0.065	3.1	2.8	c
L13	0.339	0.031	2	HG-AAS	0.0155	0.3	0.9	a
L14	0.327	0.049	2	HPLC-ICP-MS	0.0245	0.1	0.2	a
L15	0.248	0.062	2	HG-AAS	0.031	-1.2	-2.2	a
L17	0.417	0.095	2	IEC-ICP-MS	0.0475	1.6	1.9	a
L18	0.328	0.043	2	ICP-MS	0.0215	0.1	0.3	a
L23	0.34	0.068	2	HPLC-ICP-MS	0.034	0.3	0.5	a
L24	0.45	0.18	2	HG-AAS	0.09	2.1	1.4	c
L26	0.428	8	2	HPLC-ICP-MS	4	1.7	0.0	c
L27	0.411	0.1644	2	ICP-MS	0.0822	1.5	1.1	c
L28	0.42	0.1	2	HPLC-ICP-MS	0.05	1.6	1.9	a
L30	0.3	0.06	2	LC ICP-MS	0.03	-0.3	-0.7	a

[a] √3 is set by the ILC coordinator when no expansion factor k is reported. The reported uncertainty was assumed to have a rectangular distribution with $k = √3$.

[b] **Satisfactory, Questionable, Unsatisfactory**

[c] **a** : u_{min} (u_{ref}) ≤ u_{lab} ≤ u_{max} (σ_p);   **b** : u_{lab} < u_{min};   and **c** : u_{lab} > u_{max} (σ_p)

IMEP-116: Inorganic arsenic in mushroom

X_{ref} = 0.321; U_{ref} = 0.026 (k=2); σ_p = 0.06 (mg kg^{-1})
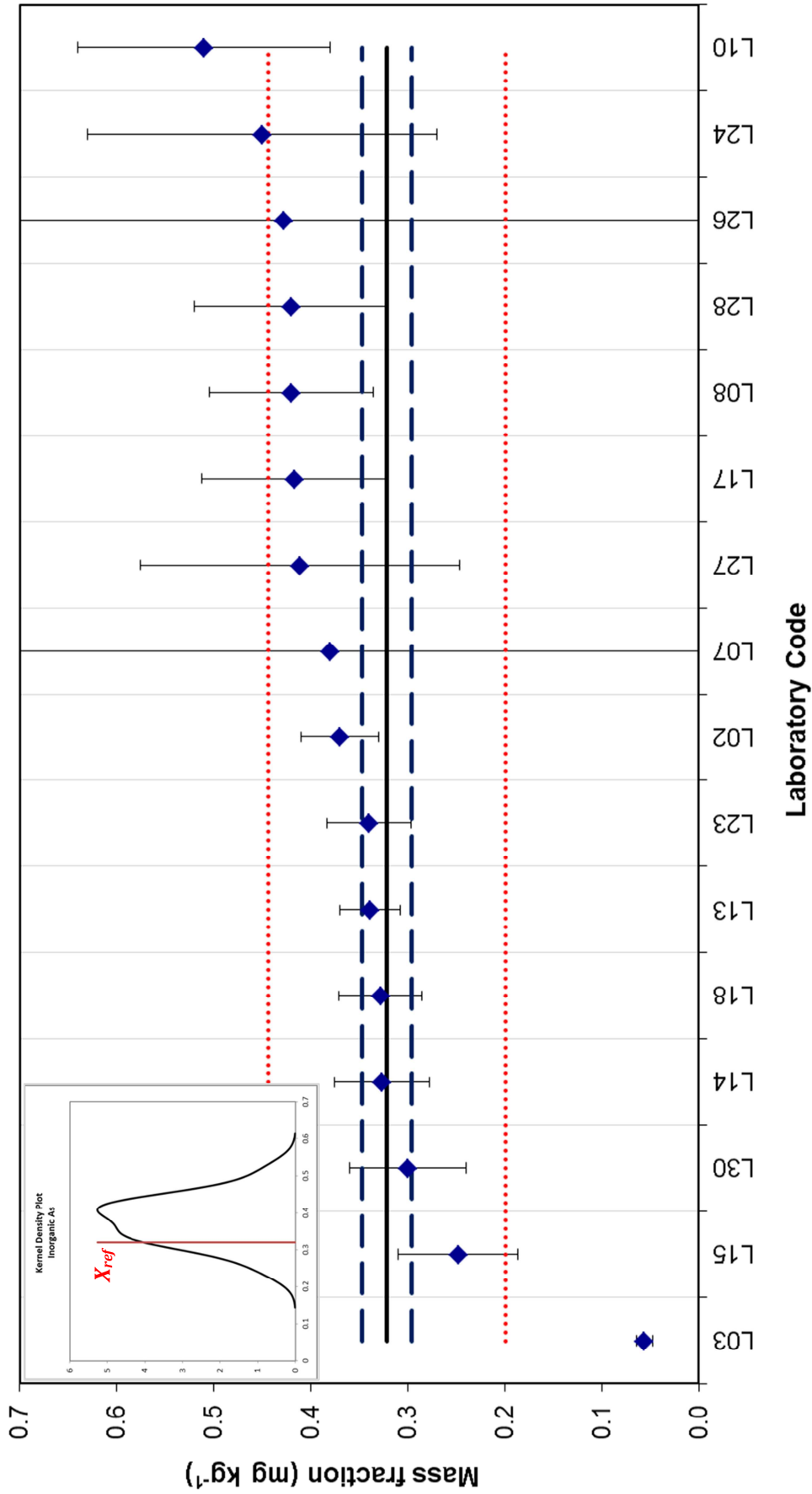
Measurement results and associated uncertainties (reported uncertainties shown).
Reference value (X_{ref}): solid black line; Reference interval ($X_{ref} \pm U_{ref}$): dashed blue lines; Target interval ($X_{ref} \pm 2\sigma_p$): dotted red lines.

Annex 12: Experimental details and scoring

(cf. Annex 5, questions Q2.1, Q08, Q09, Q11, Q12.1 and Q16)

Lab. ID	Official method	CRM used	Digestion	Digestion acids	Technique	LoD (mg kg^{-1})	Analysis frequency	z-scoring
L01								Total As
	EN 14084	NIST1568a rice flour	Microwave	HNO$_3$ + H$_2$O$_2$	ET-AAS	0.010	0-50	Total Cd
								Total Hg
	EN 14084	NIST 1568a rice flour	Microwave	HNO$_3$ + H$_2$O$_2$	ET-AAS	0.01	0-50	Total Pb
								iAs
L02	No				ICP-MS	0.001	0-50	Total As
		BCR Rye grass	Microwave	HNO$_3$	ICP-MS	0.001	0-50	Total Cd
					ICP-MS	0.001	0-50	Total Hg
							0-50	Total Pb
		NIST1586a		HNO$_5$	HPLC-ICP-MS	0.01	0-50	iAs
L03	EN 14546	NIST 1568	Dry mineralization		HG-AAS	0.10	50-250	Total As
	EN 14083		Microwave		ET-AAS	0.005	50-250	Total Cd
	in house	BCR 422, NIST 1566, 1568	No	HNO$_3$ + H$_2$O$_2$	DMA	0.0002	50-250	Total Hg
	EN 14083	NIST 1566, 1568	Microwave		ET-AAS	0.05	50-250	Total Pb
	in house	proficiency test leftover	Dry mineralization		HG-AAS	0.15	50-250	iAs
L04								Total As
	AOAC 999.10	FAPAS 7152	Wet digestion	HNO$_3$ + H$_2$O$_2$	AAS		50-250	Total Cd
								Total Hg
	AOAC 999.10	FAPAS 7152	Wet digestion	HNO$_3$ + H$_2$O$_2$	AAS		50-250	Total Pb
								iAs
L05	EN 16206				HG-AAS	0.050	50-250	Total As
	EN 15550	BRC 191			GF-AAS	0.050	50-250	Total Cd
			Microwave	HNO$_3$/H$_2$O$_2$	CV-AAS	0.050	50-250	Total Hg
	EN 15550	BRC 191			GF-AAS	0.050	50-250	Total Pb
							0-50	iAs
L06	EN 15763	DORM-3			ICP-MS	0.005	0-50	Total As
	EN 15763	BCR-191			ICP-MS	0.003	50-250	Total Cd
	EN 15763	DORM-3	Microwave	HNO$_3$/H$_2$O$_2$	ICP-MS	0.01	0-50	Total Hg
	EN 15763	BCR-191			ICP-MS	0.015	50-250	Total Pb
								iAs
L07	No				ICP-MS	0.0084	> 1000	Total As
		NIST SRM 1568a	Microwave	HNO$_3$	ICP-MS	0.0023	> 1000	Total Cd
					ICP-MS	0.0017	> 1000	Total Hg
					ICP-MS	0.0055	> 1000	Total Pb
		Rice ERM-BC211			HPLC-ICP-MS	0.03	> 1000	iAs
L08	EN 15763				ICP-MS	0.010	50-250	Total As
	EN 15763	Durum wheat flour NIST 84		HNO$_3$ + HCL	ICP-MS	0.003	50-250	Total Cd
	EN 15763		Microwave		ICP-MS	0.019	50-250	Total Hg
	EN 15763				ICP-MS	0.004	50-250	Total Pb
		Rice NMIJ 7503-a		HNO$_3$ + H$_2$O$_2$	ICP-MS	0.0025	0-50	iAs

Lab. ID	Official method	CRM used	Digestion	Digestion acids	Technique	LoD (mg kg^{-1})	Analysis frequency	z-scoring
L09	No							Total As
		BCR-191 and BCR-610	Microwave	HNO$_3$	ET-AAS	0.003	0-50	Total Cd
								Total Hg
		BCR-191 and BCR-713	Microwave	HNO$_3$	ET-AAS	0.008	0-50	Total Pb
								iAs
L10	No		Microwave oven	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.005		Total As
			Microwave oven	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.005		Total Cd
		BCR, NIST	No pretreatment		DMA	0.010		Total Hg
			Microwave oven	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.005		Total Pb
				HNO$_3$	LC-ICP-MS			iAs
L11	EN 15763			HNO$_3$ + H$_2$O$_2$	ICP-MS	0.020	0-50	Total As
	EN 15763			HNO$_3$ + H$_2$O$_2$	ICP-MS	0.001	50-250	Total Cd
	EPA 7473	NCS ZC73012	Microwave		DMA	0.005	0-50	Total Hg
	EN 15763			HNO$_3$ + H$_2$O$_2$	ICP-MS	0.010	50-250	Total Pb
								iAs
L12	No				ET-AAS	0.85	50-250	Total As
		TOMATO LEAVES NIST 1573a	Microwave	HNO$_3$ + H$_2$O$_2$ + HF	ET-AAS	0.25	50-250	Total Cd
					DMA	0.034	50-250	Total Hg
					ET-AAS	1.80	50-250	Total Pb
								iAs
L13	EN 14546		Ashing using HNO$_3$		HG-AAS	0.04		Total As
			Open wet using HNO$_3$		ET-AAS	0.006		Total Cd
				HNO$_3$	CV-AAS	0.04		Total Hg
					ET-AAS	0.08		Total Pb
			Ashing using HNO$_3$		HG-AAS	0.06		iAs
L14	No		Microwave		ICP-MS		50-250	Total As
		GBW07602, 07603, Astasol			ICP-MS		50-250	Total Cd
				HNO$_3$	DMA		50-250	Total Hg
			Microwave		ICP-MS		50-250	Total Pb
					HPLC-ICP-MS		0-50	iAs
L15	EN 15763				ICP-MS	0.025	> 1000	Total As
	EN 15763				ICP-MS	0.006	> 1000	Total Cd
	EN 13806	NIST 1547	Microwave	HNO$_3$	CV-AAS	0.0025	> 1000	Total Hg
	EN 15763				ICP-MS	0.02	> 1000	Total Pb
	EN 16278				HG-AAS		0-50	iAs
L16			Dry		HG-AAS	0.010		Total As
	AOAC 99		Dry		ET-AAS	0.010		Total Cd
	AOAC 97		Wet		CV-AAS	0.002		Total Hg
	AOAC 99		Dry		ET-AAS	0.010		Total Pb
								iAs
L17	No				ICP-MS	0.0072	50-250	Total As
			Microwave, wet pressure		ICP-MS	0.0015	50-250	Total Cd
		NIST 2976		HNO$_3$	ICP-MS	0.0072	50-250	Total Hg
					ICP-MS	0.0043	50-250	Total Pb
		BC 211	No		IEC-ICP-MS	0.010	50-250	iAs

Lab. ID	Official method	CRM used	Digestion	Digestion acids	Technique	LoD (mg kg^{-1})	Analysis frequency	z-scoring
L18	No	BCR 185r	Microwave	HNO$_3$ + HCL	ICP-MS	0.01	> 1000	Total As
					ICP-MS	0.0004	> 1000	Total Cd
		NIST 8414			ICP-MS	0.007	> 1000	Total Hg
		BCR 185r			ICP-MS	0.006	> 1000	Total Pb
					ICP-MS	0.016	50-250	iAs
L19	EN 13806							Total As
	EN 15763	DORM 3, DORM 4, MR	Microwave	3 ml HNO$_3$ + 1 ml H$_2$O$_2$	ICP-MS	0.08	0-50	Total Cd
					CV-AAS	0.003	0-50	Total Hg
	EN 15763				ICP-MS	0.52	0-50	Total Pb
								iAs
L20	No		Open wet	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.001	0-50	Total As
			Open wet		ICP-MS	0.0005	0-50	Total Cd
		BCR 186	Automatic analyser		CV-AAS	0.0004		Total Hg
			Open wet		ICP-MS	0.002	0-50	Total Pb
								iAs
L21	EN 15763	LGC 7161	Microwave	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.03	0-50	Total As
	EN 14084	LGC 7162			ET-AAS	0.0021	0-50	Total Cd
	ASU L0019/4	NRC TORT2			CV-AAS	0.005	0-50	Total Hg
	EN 14084	LGC 7162			ET-AAS	0.012	0-50	Total Pb
								iAs
L22	No	LGC 7162	Microwave	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.10	0-50	Total As
					ICP-MS	0.01	0-50	Total Cd
		NRC TORT2			ICP-MS	0.02	0-50	Total Hg
		LGC 7162			ICP-MS	0.06	0-50	Total Pb
								iAs
L23	No	NIST 1570a	Microwave	HNO$_3$ + H$_2$O$_2$	ICP-MS		> 1000	Total As
					ICP-MS		> 1000	Total Cd
					DMA		> 1000	Total Hg
					ICP-MS		> 1000	Total Pb
					HPLC-ICP-MS			iAs
L24	No	internal ref.	Microwave	HNO$_3$	ICP-AES		> 1000	Total As
					ET-AAS		> 1000	Total Cd
			no		DMA		> 1000	Total Hg
				HNO$_3$	ET-AAS		> 1000	Total Pb
		FAPAS	no		HG-AAS		0-50	iAs
L25	No		Microwave	HNO$_3$	ET-AAS	0.05		Total As
					ET-AAS	0.002		Total Cd
					CV-AAS	0.01		Total Hg
					ET-AAS	0.01		Total Pb
								iAs
L26	EPA	Yes	Microwave	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.006	> 1000	Total As
	EPA				ICP-MS	0.015	> 1000	Total Cd
	AOAC				DMA	0.00012	> 1000	Total Hg
	EPA				ICP-MS	0.022	> 1000	Total Pb
		NO			HPLC-ICP-MS	0.07	0-50	iAs

Lab. ID	Official method	CRM used	Digestion	Digestion acids	Technique	LoD (mg kg^{-1})	Analysis frequency	z-scoring
L27	NMKL 186; 20	Oyster Tissue and Tort-2	Microwave	HNO$_3$ + H$_2$O$_2$	ICP-MS		> 1000	Total As
	NMKL 186; 20				ICP-MS		> 1000	Total Cd
	NMKL 186; 20				ICP-MS		> 1000	Total Hg
	NMKL 186; 20				ICP-MS		> 1000	Total Pb
		BCR 627 and Tort-2			ICP-MS		50-250	iAs
L28	No	IRMM-804	Microwave	HNO$_3$	ICP-MS		> 1000	Total As
					ICP-MS		> 1000	Total Cd
		BCR-150, NIST 2976			DMA		> 1000	Total Hg
		IRMM-804	Microwave	HNO$_3$	ICP-MS		> 1000	Total Pb
		NMIJ 7503a	Microwave	HNO$_3$ + H$_2$O$_2$	HPLC-ICP-MS		0-50	iAs
L29	No	ERM-CD 281	Microwave		ICP-MS	0.05		Total As
					ICP-MS	0.02		Total Cd
					ICP-MS	0.1		Total Hg
					ICP-MS	0.2		Total Pb
								iAs
L30	No	BCR 482	Microwave	HNO$_3$	ET-AAS			Total As
					ET-AAS			Total Cd
					CV-AAS			Total Hg
					ET-AAS			Total Pb
					LC-ICP-MS			iAs
L31	EN 15763		Microwave	HNO$_3$	ICP-MS	0.00005	50-250	Total As
	EN 15763				ICP-MS	0.00001	50-250	Total Cd
					DMA	0.008	50-250	Total Hg
	EN 15763				ICP-MS	0.00002	50-250	Total Pb
								iAs
L32	EN 15763		Microwave	HNO$_3$	ICP-MS	0.010		Total As
	EN 15763				ICP-MS	0.002	50-250	Total Cd
	EN 15763				ICP-MS	0.010		Total Hg
	EN 15763				ICP-MS	0.005		Total Pb
								iAs
L33			Dry ashing	Mg(NO$_3$)$_2$	AAS	0.050	50-250	Total As
			Dry ashing	Mg(NO$_3$)$_3$	AAS	0.100	50-250	Total Cd
			Wet digestion	HNO$_3$ + H$_2$SO$_4$	CV-AAS	0.005	50-250	Total Hg
			Dry ashing	Mg(NO$_3$)$_2$	AAS	0.5	50-250	Total Pb
								iAs
L34	AOAC 90, N°3		Microwave	5 ml HNO$_3$+3ml H$_2$O$_2$	ICP-MS	0.003	0-50	Total As
	AOAC 999.10				AAS	0.003	50-250	Total Cd
	AOAC 90, N°3				ICP-MS		50-250	Total Hg
	AOAC 999.10				AAS	0.014	50-250	Total Pb
								iAs
L35	EN 14627	NIST 1515	Microwave	HNO$_3$ + H$_2$O$_2$	HG-AAS	0.200	0-50	Total As
	EN 14084				ET-AAS	0.010	50-250	Total Cd
	EN 13806				CV-AAS	0.050	0-50	Total Hg
	EN 14084				ET-AAS	0.020	50-250	Total Pb
								iAs

Lab. ID	Official method	CRM used	Digestion	Digestion acids	Technique	LoD (mg kg^{-1})	Analysis frequency	z-scoring
L36					ICP-MS			Total As
					ICP-MS			Total Cd
					ICP-MS			Total Hg
					ICP-MS			Total Pb
								iAs
L37	No				ICP-MS	0.015	0-50	Total As
			Microwave	HNO$_3$ + H$_2$O$_2$	ICP-MS	0.010	0-50	Total Cd
					ICP-MS	0.019	0-50	Total Hg
					ICP-MS	0.015	0-50	Total Pb
							0-50	iAs

Abstract

This report presents the results of a proficiency test exercise (PT) focussed on the determination of total cadmium, lead, arsenic, mercury and inorganic arsenic in mushrooms. The exercise was organised in support of the EU Regulation 1881:2006 which sets the maximum levels for certain contaminants in foodstuffs.

Thirty eight participants from twenty six countries registered to the exercise. Only one participant did not report results.

The test item used was a blend of mushooms (of the variety Lentinula edodes). The assigned value was obtained as the average of results reported by five expert laboratories having demonstrated experience in the analysis of trace elements in different matrices. The associated uncertainties of the assigned values were computed according to the ISO/IEC Guide 98:2008 (GUM) and following ISO 13528:2005.

Participants were invited to report their measurement uncertainties. Laboratory results were rated with z- and zeta (ζ-) scores in accordance with ISO 13528:2005. The standard deviation for the proficiency assessment was based on the use of the modified Horwitz equation (for inorganic arsenic (19 % of X_{ref}) and for the total mass fraction of lead, 20 % of X_{ref}) while slightly lower percentages were decided, upon expert judgment of the advisory board of this PT exercise and based on previous participant's performance on similar measurands, for the total mass fractions of arsenic and mercury (15 % of X_{ref}) and 10 % of X_{ref} for the total mass fraction of cadmium.

The percentage of satisfactory z-scores ranged from 81 % (inorganic arsenic) to 97 % (total cadmium). Therefore, the outcome of the exercise shows an overall excellent performance for European National Reference Laboratories assuring compliance towards the European legislation related to the determination of the investigated food contaminants.

As the Commission's in-house science service, the Joint Research Centre's mission is to provide EU policies with independent, evidence-based scientific and technical support throughout the whole policy cycle.

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Key policy areas include: environment and climate change; energy and transport; agriculture and food security; health and consumer protection; information society and digital agenda; safety and security including nuclear; all supported through a cross-cutting and multi-disciplinary approach.

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